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Dynamics of plum curculio, Conotrachelus nenuphar (Herbst.) (Coleoptera: Curculionidae) : immigration into apple orchards.

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DYNAMICS OF PLUM CURCULIO, *CONOTRACHELUS NENUPHAR* (HERBST.) (COLEOPTERA: CURCULIONIDAE), IMMIGRATION INTO APPLE ORCHARDS

A Dissertation Presented

by

JAIME CESAR PIÑERO

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DEDICATION

To Ron J. Prokopy, for all his guidance, support, motivation, and friendship

To Isabel, for her love, patience, and inspirational way of thinking

To my mother, with infinite love and vast gratitude

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ABSTRACT

DYNAMICS OF PLUM CURCULIO, *CONOTRACHELUS NENUPHAR* (HERBST.) (COLEOPTERA: CURCULIONIDAE), IMMIGRATION INTO APPLE ORCHARDS

MAY 2005

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This project focused on field evaluations of synthetic host and pheromonal odor combinations for attractiveness to plum curculios, *Conotrachelus nenuphar* (Coleoptera: Curculionidae), with the aim of developing a monitoring device that could track the onset and extent of immigration into apple orchards, thereby maximizing plum curculio control while minimizing exposure and cost by helping growers to accurately time insecticide sprays. In a 2000 field test I screened six individual host plant volatiles, each in association with grandisoic acid (synthetic plum curculio aggregation pheromone), using panel and pyramid traps. Benzaldehyde was the most attractive host plant odor when in combination with grandisoic acid. Evaluations conducted in 2001 confirmed the attractiveness of this binary combination to both males and females across the entire period of immigration, and also revealed that benzaldehyde interacts in a synergistic manner with grandisoic acid. Additional field studies conducted in 2002 and 2003

showed that benzaldehyde at 40 mg/day of release in association with grandisoic acid at 1 mg/day of release is the most cost-effective bait combination for use in traps to monitor plum curculio immigration.

I also investigated temporal dynamics of plum curculio immigration into an unsprayed section of an apple orchard over a five-year period using traps. My main goal was to determine relationships among the timing of immigration, temperature and phenological tree stage. Based on my data, I propose a pre- and a post-petal-fall period of immigration, each of which is influenced to a different extent by spring temperature. Thermal constants (expressed in Degree Days [DD] base 6.1°C, computed starting January 1st) for the start of immigration, 50th and 80th percentile of cumulative captures were 113, 249 and 412 DD, respectively.

Some spatial aspects of plum curculio immigration were also studied in commercial apple orchards over a two-year period. By petal fall, most adults were found on perimeter-row trees in sprayed blocks having large trees (M.7 rootstock), but more adults were found inside of blocks than on perimeter-row trees if blocks had small trees (M.9 rootstock). Plum curculios were able to overwinter inside blocks, regardless of the presence or absence of weeds beneath trees.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

INTRODUCTION

Apple (*Malus domestica* Borkh) is the most economically valuable tree-fruit crop in eastern North America. The value of apple production combining all states in the Northeast (CT, DE, MA, MD, ME, NJ, NH, NY, PA, RI, VT, and WV) was estimated in nearly $338 million in 2003 (NASS-USDA 2004). Apple production in eastern North America, however, is threatened every year by a complex of more than 22 insect pests and 8 diseases (ProNew England Apple Crop Profile 2003). Among all insect pests associated with apple, the plum curculio, *Conotrachelus nenuphar* (Herbst) (Coleoptera: Curculionidae), stands as one of the most devastating in eastern North America (Racette et al. 1992).

The plum curculio is native to North America where, before the introduction of cultivated fruits, it reproduced mainly on host plants of the genus *Prunus* (wild plums) (Whitcomb 1929). As early as 1736, it was reported as a serious pest of garden plums in some areas of the northeast (Quaintance and Jenne 1912). Currently, the plum curculio attacks nearly all stone and pome fruit (plant family: Rosaceae) with plum, apple, cherry, pear, peach, nectarine, and apricot as its preferred hosts (Racette et al. 1992). However, other plant species such as high-bush blueberry (plant family: Ericaceae) may also be attacked by plum curculio (Chen 1965).

Regarding its geographic distribution, the plum curculio is widespread east of the Rocky Mountains, with southern Canada and the Gulf of Mexico as the latitudinal limits

(Whitcomb 1929). In the northern part of its range, the plum curculio completes only one generation per year (i.e., it is univoltine), with adults overwintering in a state of obligatory diapause (Chapman 1938, Smith 1957). South of Virginia, however, there are usually two generations per year (i.e., it is bivoltine), with adults undergoing a period of apparent facultative diapause during the winter months (Schoene 1936, Smith 1957). There seems to be some degree of reproductive incompatibility between the northern univoltine strain and the southern bivoltine strain (Padula and Smith 1971). In addition, the supercooling points (Ch. Vincent, unpublished) and the behavioral responses of northern-strain plum curculios to semiochemicals also appear to be different from those exhibited by southern-generation plum curculios (Leskey and Wright 2004). The degree of differentiation between northern and southern populations of plum curculios has been studied in more detail by McClanan et al. (2004) using molecular approaches.

The life history of the plum curculio is very well known, as it has been worked out carefully by many researchers. In brief, the plum curculio passes through four stages of development: egg, larva, pupa and adult. The life cycle starts when an overwintered adult female lays individual eggs under the skin of the developing host fruit, first using her mouthparts to cut a small crescent shaped flap in the fruit skin and then turning around to deposit an egg (Quaintance and Jenne 1912). The duration of each stage is highly dependent on prevailing temperatures (Quaintance and Jenne 1912, Chapman 1938). Within 4-11 days of being laid, larvae hatch from the eggs and tunnel into the flesh of the fruit as they feed. Larvae complete four instars inside the fruit in about 16 days. Pupation occurs in the soil and lasts between 4-5 weeks. For the northern strain, upon emergence from pupae during the summer, adults feed on fruit but do not mate.

Subsequently, summer-generation adults overwinter, and then re-appear in host trees the following spring. In Connecticut and Massachusetts, approximately 55 days are required by the plum curculio to develop from a newly laid egg to an adult, depending on prevailing temperatures (Garman and Zappe 1929, Whitcomb 1929).

Since the first report published by Riley (1871), numerous studies have been conducted on plum curculio ecology and behavior (reviewed by Quaintance and Jenne 1912, Holloway 1977, Racette et al. 1992 and Vincent et al. 1999). Most work has addressed the timing of spring emergence and the within-orchard dispersal and patterns of behavior of adult plum curculios in an attempt to use this information to develop reliable monitoring and/or control methods. It has been generally agreed that in autumn (apparently from mid-September to early October under Massachusetts conditions [Withcomb 1929]), most adults in northeastern North America seek refuge in woods or hedgerows and overwinter there in a state of dormancy (diapause), although some plum curculios are capable of overwintering within orchard blocks if there is grass or accumulations of leaves (Chapman 1938, Lafleur et al. 1987). Winter mortality of plum curculio adults has been reported to be as low as 23% (Smith and Flessel 1968) or as high as 100% (Whitcomb 1929, Smith and Flessel 1968), depending on weather prevailing during the winter months. The main factors responsible for such high levels of mortality of the diapausing adults seem to be desiccation and disruption of tissue caused apparently by lack of moisture, poor snow cover, and excessively low temperatures in December, January and February (Quaintance and Jenne 1912, Lathrop 1949). Depletion of lipid reserves caused by excessively high temperatures experienced by early diapausers may be another important mortality factor (Danks 1987, Leather et al. 1995).

The timing of emergence of plum curculios from overwintering habitats and their immigration into apple orchards has been related primarily to soil and air temperatures and to host plant phenology. However, the relative influence of these two types of environmental factors on timing and extent of plum curculio immigration has not been determined. Temperature has been referred as to the primary environmental factor triggering plum curculio emergence and immigration into orchard trees by most researchers (e.g., Quaintance and Jenne 1912, Chapman 1938, Bobb 1949, Smith and Flessel 1968, McGiffen and Meyer 1986). For example, Smith and Flessel suggested a 21°C requirement (air temperature) for adult emergence, and Quaintance and Jenne (1912) found that a mean air temperature ranging between 12.8°C and 15.6°C for 3-4 days was required to induce migration to host trees. A soil temperature of 10°C (at 7.6 cm in depth) was proposed by Bobb (1949) as the threshold for adult emergence from overwintering sites. In contrast, others (e.g., Whitcomb 1929, Armstrong 1958) related the seasonal abundance of plum curculios to phenological tree stage.

Because colonization of host trees by adult plum curculios in the spring may occur before, during, or after blossoming, feeding may occur first on the buds, flowers and, subsequently, newly set fruit (Smith and Flessel 1968, Lafleur and Hill 1987). In Quebec and other areas of the northeast (e.g., New York, Massachusetts) the highest population densities of plum curculio adults in apple orchards have been recorded 1-2 weeks after petal fall (Garman and Zappe 1929, Whitcomb 1929, Chapman 1938, Lathrop 1949, Racette et al. 1992). However, seasonal monitoring of adult activity on host trees has been determined almost exclusively by tree-limb jarring because no other monitoring technique has been available. This approach involves tapping limbs using a

pole to dislodge plum curculio adults onto an underlying ground cloth (Quaintance and Jenne 1912, Chapman 1938).

Adult plum curculios may arrive to host trees by either direct flight or by crawling (Blanchett, 1987, Prokopy et al. 1999), depending on weather (Dixon et al. 1999). Upon reaching orchard trees, plum curculios have shown a tendency to stay in perimeter-row trees (Quaintance and Jenne 1912, Rings 1952), but as the season progresses plum curculios may move from perimeter-row trees toward interior trees (LaFleur and Hill 1987). Whether tree size is a factor influencing the proclivity of plum curculios to stay on perimeter-row trees or penetrate into interior trees is an aspect that has not been evaluated.

The level of plum curculio activity and degree of within-orchard dispersal once adults are present on host trees has been related largely to phenological tree stage. Whereas some authors (e.g., Snapp 1940, Rings 1952, Chouinard et al. 1992a) report little inter-tree movement of plum curculio adults, others (e.g., Lathrop 1949, Smith and Flessel 1968) consider that adults do not stay on host trees continuously but instead redistribute themselves on a daily basis, even during favorable conditions.

Activity patterns of plum curculio are largely crepuscular (Smith and Flessel 1968, Owens et al. 1982, Racette et al. 1990, Chouinard et al. 1993). The different types of behavior exhibited by adults (e.g., crawling, mating) depend primarily upon host plant phenology, time of day, and weather factors (Chouinard et al. 1993). For instance, the level of activity (e.g., feeding, flight, movement leading to aggregations) of plum curculio adults is much higher when trees are at bloom than at petal fall or fruit set, once the requirements for feeding and mating have been met (Racette et al. 1990, 1991, 1992,

Chouinard et al. 1993). When fruit reach 6-7 mm diameter, they become susceptible for oviposition (Chapman 1938). Levine and Hall (1977) reported that pectic enzymes and cellulose, released by the larvae as they feed on the fruit, are responsible for premature abscission of a significant proportion of fruit damaged by plum curculio, particularly before fruit reaches 3 cm in diameter. Apples with egglaying scars that remain on the trees become deformed as they mature, thereby causing important economic losses to growers (Racette et al. 1992).

Attraction of adult plum curculios to their host plants involves, as with most other phytophagous insects, both olfactory and visual elements of behavior (Bernays and Chapman 1994). However, how adults locate essential resources such as feeding and oviposition sites is not well understood. Work by Butkewich and Prokopy (1993, 1997) conducted both in the laboratory and in the field has revealed that adult plum curculios respond positively to olfactory, visual and tactile cues. More recently, it was determined that adults respond positively not only to visual stimuli represented by trunk-mimicking (Prokopy and Wright 1998) and branch-mimicking (Leskey and Prokopy 2002) traps, but also to a synthetic male-produced aggregation pheromone (Eller and Bartelt 1996) as well as to particular volatiles emitted by host fruit (Leskey and Prokopy 2000, 2001, Prokopy et al. 2001, Leskey et al. 2001).

The development of an effective trap for monitoring or controlling an insect pest ultimately depends on a comprehensive understanding of the behavior of the pest (Foster and Harris 1997). For instance, an important aspect of pest behavior is the manner by which pest individuals approach natural resources, namely food, mates or egg-laying sites and the types of stimuli eliciting such behaviors. In this regard, Prokopy et al. (1999) and

Dixon et al. (1999) reported that plum curculios are more prone to enter tree canopies by flight rather than by walking when temperatures are above 20°C, whereas crawling seems to be the main way of approaching host trees at lower temperatures.

For effective management of plum curculio in commercial apple orchards, one of the greatest challenges has been to determine the need and timing of insecticide applications that will protect fruit from injury by adults (Racette et al. 1992, Vincent et al. 1999, Prokopy et al. 2003, 2004). In concept, a reduction in the amount of insecticide used against plum curculio, from the current norm of 3 spray applications during May and June to an amount that is precise, according to need should be accompanied with an effective approach to monitoring the course of plum curculio immigration. In particular, one of the biggest challenges has been establishing the relationship between the seasonal life history of the plum curculio and the timing of spray applications in commercial apple orchards. Among the methods traditionally used to determine the time of first appearance, location and relative abundance of adult plum curculios within an orchard, is limb jarring (Quaintance and Jenne 1912, Chapman 1938, Racette et al. 1992). However, limb jarring has several shortcomings since it (1) is labor intensive; (2) can dislodge blossoms or fruit, (3) is not very accurate, as its efficacy is dependent upon tree size, weather and other factors; (4) cannot be performed at night, the time of day at which plum curculios are most active on trees (Racette et al. 1991; Chouinard et al. 1992a, 1993); and (5) cannot be used to study immigration because plum curculios that successfully overwinter beneath perimeter-row trees will be confounded with true immigrants.

Developing a monitoring device that is efficient at attracting and capturing plum curculios to determine time of first appearance and abundance involves not only the

identification of synthetic host plant volatiles that are attractive to adults, but also exploitation of visual cues that aid adult plum curculios in locating host plants (Leskey and Prokopy 2002). Thus, the main objectives of this dissertation were, first, to develop a monitoring device that could track accurately the onset and extent of plum curculio immigration into apple orchards in early spring, and second, by using odor-baited traps, to investigate spatio-temporal aspects of plum curculio immigration. In particular, my approach involved the use of four different trap types: panel, pyramid, Circle and emergence traps. Chapters 2 and 3 present the results of evaluations of several synthetic host plant odors conducted under field conditions from 2000-2003. Chapter 4 addresses temporal dynamics of plum curculio immigration into an unsprayed section of a commercial apple orchard using odor-baited traps in an attempt to establish the relationships among the timing of immigration, temperature, and phenological tree stage. My approach included the estimation of thermal constants, expressed in Degree Days (base 6.1°C and computed starting January 1st), for different stages of plum curculio immigration. Chapter 5 reports on the early-season distribution of adult plum curculios within orchard blocks in Massachusetts, as well as the determination of not only the extent to which adults can overwinter within orchard blocks, but also the degree of plum curculio penetration into orchard blocks early in the spring.

CHAPTER 2

PLUM CURCULIO (COLEOPTERA: CURCULIONIDAE) RESPONSE TO ODOR-BAITED TRAPS NEAR WOODS

Abstract

I investigated the response of overwintered plum curculios, *Conotrachelus nenuphar* (Herbst), to odor-baited traps from the beginning until nearly the end of emigration from overwintering sites in woods. I utilized clear sticky Plexiglas panels and black pyramid traps placed close to woods adjacent to apple trees in an unsprayed section of an orchard. Traps were baited with aggregation pheromone (grandisoic acid) alone or in combination with one of six synthetic fruit volatiles (benzaldehyde, decyl aldehyde, *E*-2-hexenal, ethyl isovalerate, hexyl acetate, or limonene). Unbaited traps served as a control treatment. Plum curculio emigration from woods was divided into early-, mid-, and late-season periods based primarily on phenological stage of apple bud and fruit development (tight cluster to bloom, petal fall, and fruit set, respectively). During both early- and late-season, panel and pyramid traps baited with benzaldehyde plus pheromone were significantly more attractive than any other traps (baited or unbaited), except panel traps baited with ethyl isovalerate plus pheromone in early season, which likewise captured significantly more adults than unbaited panel traps. During mid-season, no lures were significantly attractive, possibly due to prevailing cool weather, unfavorable for adult activity. Over the entire season, panel or pyramid traps baited with benzaldehyde

plus pheromone captured nearly 6 times as many plum curculios as unbaited traps of each type, whereas traps baited with pheromone alone captured about twice as many as unbaited traps of each type. I conclude that panel or pyramid traps, or a combination, baited with benzaldehyde plus pheromone placed at borders of plum curculio overwintering sites can be a valuable tool for monitoring the beginning, peak, and end of adult immigration into apple orchards.

Introduction

The plum curculio, *Conotrachelus nenuphar* (Herbst) (Coleoptera: Curculionidae), is a major pest of stone and pome fruit in eastern and central North America (Racette et al. 1992). Most plum curculio adults that attack fruit in orchards during spring immigrate from overwintering sites outside the orchard, or near the edges, depending on hibernation site characteristics (Lafleur and Hill 1987). In the absence of an effective trap to aid in determining extent and timing of immigration and hence, need and timing of insecticide treatment, growers annually apply two or three sprays of azinphosmethyl, phosmet, or similar insecticide to prevent damage inflicted by this insect (Anonymous 1997, Reissig et al. 1998). Because early warning of plum curculio population density and activity level could forestall insecticide treatment until needed, recent studies have focused on development of baited monitoring traps to provide early warning. Both visual (e.g., tree structure mimicking) and olfactory (e.g., host plant volatile, aggregation pheromone) stimuli could be valuable components of plum curculio monitoring devices (Butkewich and Prokopy 1993, 1997; Prokopy et al. 1995).

Trap types evaluated to date for monitoring plum curculio include conical colored and rectangular white cardboard sticky traps (Le Blanc 1982), scout-apples (Le Blanc et al. 1984), green-painted plastic sticky spheres (Yonce et al. 1995), Circle traps (Mulder et al. 1997), and black pyramids, black cylinders, and vertical clear Plexiglas panels (Mulder et al. 1997, Prokopy and Wright 1998, Dixon et al. 1999, Prokopy et al. 1999a, 2000). Combined results from these studies indicate that black pyramid traps (mimicking tree trunks), clear Plexiglas panels (for capturing flying adults), black cylinder traps (mimicking tree branches), and Circle traps (for intercepting adults walking on tree trunks or limbs) may hold the greatest potential for use as plum curculio monitoring tools.

Plum curculio responses to synthetic aggregation pheromone (grandisoic acid [Eller and Bartelt 1996]) and host plant volatiles (Leskey and Prokopy 2000, Leskey et al. 2001, Prokopy et al. 2001) have been studied recently under laboratory and field conditions. Results have shown that some components of the odor of unripe plum or apple fruit (e.g., benzaldehyde, benzyl alcohol, decanal, *E*-2-hexenal, ethyl isovalerate, hexyl acetate, limonene) are attractive to plum curculios (Prokopy et al. 2001).

Whether a combination of host plant volatiles and aggregation pheromone enhances plum curculio captures by traps under field conditions has not been determined. Prokopy et al. (2000) found that clear Plexiglas panels baited with two synthetic fruit volatiles (ethyl isovalerate and limonene) in conjunction with pheromone, placed in proximity to woods, were significantly more attractive than unbaited traps, but no differences between baited and unbaited pyramid or cylinder traps placed in orchard trees were found, possibly because the release rate used was too high. Nevertheless, in some

other species of Curculionidae, such as the boll weevil, *Anthonomus grandis grandis* Boheman (reviewed by Hardee and Mitchel 1997) and the red sunflower seed weevil, *Smicronyx fulvus* LeConte (Roseland et al. 1990), and Nitidulidae (e.g., the dusky sap beetle, *Carpophilus lugubris* Murray [Bartelt et al. 1991, Hengchen et al. 1992] and the driedfruit beetle, *C. hemipterus* [L.] [Dowd and Bartelt 1991; Bartelt et al. 1992]), an additive or synergistic effect of aggregation pheromone and odor of host-plant tissue (or components thereof) on adult attraction has been reported.

The aim of this study was to investigate behavioral responses of overwintered adult plum curculios to clear Plexiglas panel and black pyramid traps baited with different synthetic fruit volatiles (each in combination with grandisoic acid), or grandisoic acid alone, during the period of immigration of plum curculios into an apple orchard. I also examined plum curculio captures in relation to time of day and various weather variables such as temperature, relative humidity, wind speed, rainfall, and barometric pressure.

Materials and Methods

Study Site

The study was carried out in an unsprayed section of a commercial apple orchard at the University of Massachusetts Horticultural Research Center in Belchertown, MA. Most trees were medium-sized (M.26 rootstock) and were 'McIntosh' and 'Red Delicious'. The experimental section was bordered by a mixed-species deciduous woods

(Fig. 1), except on one side, which faced an orchard access road, beyond which lay a commercial pear (*Pyrus communis* L.) orchard.

Trap Types

I evaluated two trap types (Fig. 2a): 1) black pyramid traps, originally designed by Tedders and Wood (1994) and modified by Prokopy and Wright (1998) and Prokopy et al. (1999a); and 2) clear Plexiglas sticky panels (60 by 60 cm), described and tested by Prokopy et al. (1998), Dixon et al. (1999), and Prokopy et al. (2000). Panel trap design was modified slightly: a clear Plexiglas panel was attached vertically to a wooden post (1.70 m), which was cut in half and fitted with a plastic sleeve (black-painted polyvinylchloride pipe, 30 cm in length), to permit lifting and moving of the top half of the post and attached panel while leaving the bottom half of the post secured in the ground (Fig. 2b). As in Prokopy et al. (2000), only the woods-facing side of each panel was coated with Tangletrap (Orchard Equipment Supply Company Inc., Conway, MA). Our purpose in deploying these two types of traps was to capture adults immigrating from woods primarily by crawling (pyramid traps) or flight (panel traps), although pyramid traps also capture flying plum curculios (Prokopy et al. 1999b, Dixon et al. 1999).

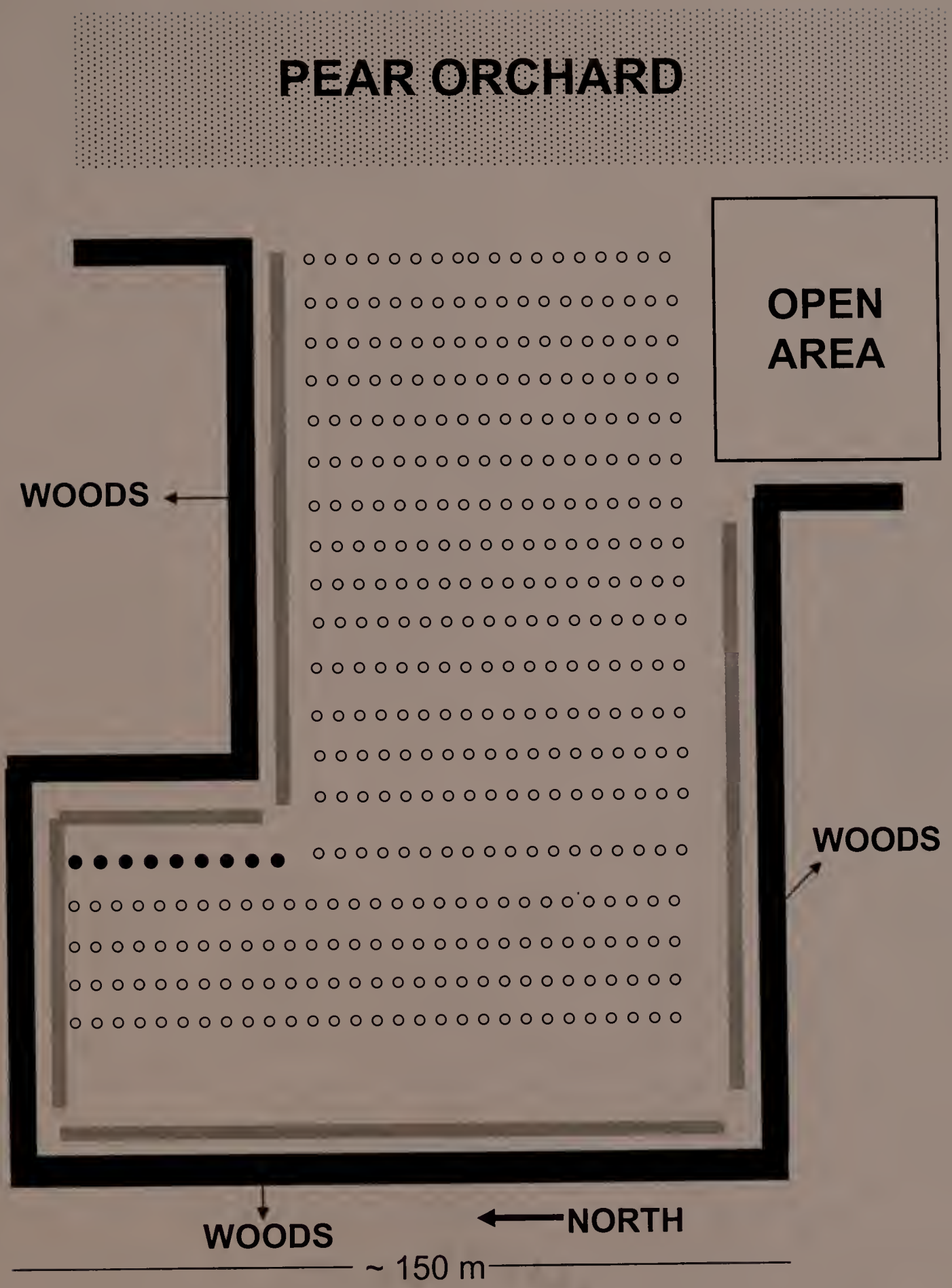

Figure 1. Diagram representing the unsprayed section of the apple orchard in which the study was carried out (Belchertown, MA). Black-filled circles represent the section of the orchard where tapping of apple tree branches for collection of adults was performed.

A

B

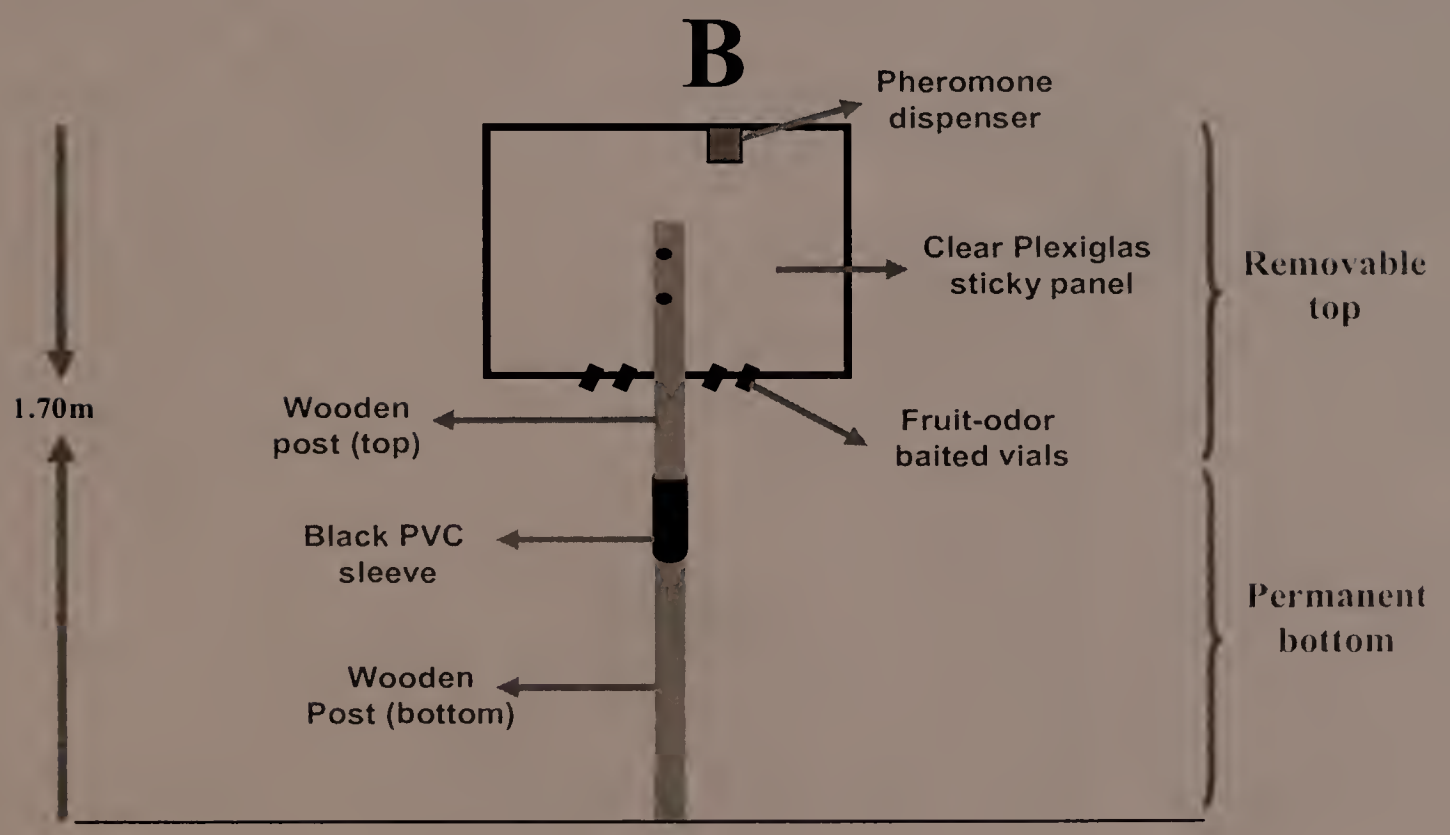

Figure 2 A) View of a pyramid and a panel trap placed in proximity to woods adjacent to the apple orchard used in the study. B) Panel trap design. Forty traps of each type were deployed for the study.

Bait Treatments

I evaluated each of six synthetic fruit volatiles in combination with grandisoic acid: benzaldehyde, decyl aldehyde, *E*-2-hexenal, ethyl isovalerate, hexyl acetate, and limonene (Aldrich Chemical Co., Milwaukee, WI). I also evaluated grandisoic acid alone and an unbaited control treatment. All six synthetic fruit volatiles had been found to elicit a positive response by plum curculios when evaluated earlier in association with boll weevil traps placed on the ground beneath host trees (Prokopy et al. 2001). Fruit volatiles were introduced into high-density polyethylene vials (400 µl) (VWR Scientific Products, Boston, MA) to facilitate controlled odor release. Different numbers of vials of each compound were needed to achieve a release rate of ~10 mg/d. Vials were attached to the lower edge of Plexiglas panels by using steel binder clips, or were placed inside the inverted screen funnel (boll weevil trap top; Gempler's, Mt. Horeb, WI) capping a pyramid trap. Pheromone lures were contained in commercial dispensers (ChemTica Internacional, S.A., San Jose, Costa Rica) that released ~1 mg of pheromone per day. A single lure was attached to the upper edge of a Plexiglas panel by using a binder clip, or was placed inside the inverted screen funnel capping a pyramid trap. Numbers of volatile-baited vials used per trap, release rates and expected duration of volatiles under laboratory conditions are given in Table 1. To maintain consistent release, vials containing benzaldehyde and limonene, as well as grandisoic acid dispensers, were replaced on 24 May because the longevity of these compounds under the conditions of this study was shorter than the longevity estimated previously in the laboratory.

Trap Setup

On 30 April 2000 (tight cluster stage of bud development), 40 clear Plexiglas panels and 40 black pyramid traps were placed along the edge of the unsprayed section of the orchard, approximately 1 m outside the foliage of woods. Traps were arranged in five groups of 16 traps each (dictated by the combination of eight odor treatments and two trap types). Each group of 16 traps represented one replicate. Traps were deployed in pairs, with one trap of each type (bearing the same odor treatment) placed 1 m apart (Fig. 2a). Each pair was separated by 10 m. Odor treatments within a replicate were assigned at random. Traps remained until 7 June (end of fruit set), when a different experiment was initiated in the same locale. This protocol revealed very little adult immigration into the orchard after 7 June.

Trap Inspection

Traps were inspected every morning (ca. 0730 hours) throughout the study. In addition, during 5-10 May, trap captures also were recorded at 1430 and 1930 hours to determine a possible temporal pattern of plum curculio response to each trap type. When trap inspection yielded five or more plum curculios across all 80 traps, I rotated (within a replicate) the top half of each panel trap and the trap atop each pyramid one position (clockwise) to minimize the influence of bait location on adult captures. In total, there were 16 rotations, which allowed each bait treatment to occupy the same location twice within a replicate. Rotations were completed on 9 May and 7 June.

Weather Information

Hourly temperature, relative humidity, and rainfall records were obtained from a CR10-X data-logger (Campbell Scientific Co., North Logan, UT) at Belchertown, MA. Wind speed was recorded through a contact anemometer (Mod. W164-B/M; Weather Measure Co., Sacramento, CA), placed in the center of the experimental plot. Barometric pressure records were provided by Skybit, Inc. (Boalsburg, PA). For the purposes of regression analyses (see below), I used mean daily values of each weather variable. Rainfall values represented total accumulation per day.

Statistical Analyses

Plum curculio immigration was divided into three seasonal time periods based on phenological stages of apple bud and fruit development and weather conditions (Fig. 3). For each time period, I performed a two-way ANOVA (trap type x treatment bait), followed by a Fisher-protected LSD separation of treatment means. For each bait treatment, I pooled, within each of the five replicates, plum curculio captures across the number of days comprising the replicate (30 April-9 May, 10-24 May, 25 May-7 June). A two-way ANOVA was performed for the comparison of trap captures according to trap type and time of day. In both analyses, the response variable was the number of adults captured by traps (in the second analysis, only by panel and pyramid traps baited with benzaldehyde plus pheromone), following square-root transformation of each value plus 0.5 to homogenize variances. In all cases, non-transformed data are presented to facilitate

interpretation. I performed stepwise linear multiple regression analyses (Darlington 1990) to determine the effect of weather conditions on male and female responses to traps baited with either benzaldehyde plus grandisoic acid or grandisoic acid alone throughout the study period.

Results

In all, 387 plum curculio adults (182 males and 205 females) were captured by traps during the 39-d study period. Early season occurred during 30 April-9 May (from tight cluster to bloom), a period characterized by high temperature (mean 18.5°C, max 32.9°C) and little rainfall (7.1 mm). During this period, 266 adults were captured, corresponding to 69% of the seasonal total (Fig. 3). Mid-season captures occurred 10-24 May (during petal-fall), a period characterized by low temperature (mean 12.3°C, max 22.3°C) and considerable rainfall (98.5 mm). During this period, 36 plum curculios were captured, corresponding to 9% of the seasonal total. Late season occurred during fruit set (25 May-7 June). This period was characterized by a combination of warm and cool days (mean 14.8°C, max 30.0°C) and little rainfall (12.0 mm). Eighty-five plum curculios (22% of the seasonal total) were caught by traps during this period.

Early Season

Regardless of bait treatment, during this period panel traps captured significantly more adults ($df = 1, 64$; $F = 9.56$; $P = 0.003$) than pyramid traps (mean ± SE, 4.15 ± 0.53

and 2.50 ± 0.50, respectively). Panel traps baited with benzaldehyde plus pheromone captured significantly more adults than any other panel traps except those baited with ethyl isovalerate plus pheromone, which captured significantly more adults than unbaited panel traps but not significantly more than other baited panel traps (df = 7, 64; F = 2.36; $P = 0.03$) (Fig. 4). No other baited panel traps captured significantly more adults than unbaited traps. Pyramid traps baited with benzaldehyde plus pheromone captured significantly more adults than any other pyramid traps, followed by traps baited with pheromone alone, which captured significantly more adults than unbaited pyramid traps (df = 7, 64; F = 5.0; $P = 0.0001$) (Fig. 4). No other baited pyramid traps captured significantly more adults than unbaited traps.

Mid Season

During this period, no significant differences were found in numbers of adults captured by panel versus pyramid traps (df = 1, 64; F = 0.43; $P > 0.05$) (mean ± SE, 0.50 ± 0.11 and 0.40 ± 0.10, respectively). Also, for neither panel (df = 7, 64; F = 0.87; $P > 0.05$) nor pyramid (df = 7, 64; F = 0.57; $P > 0.05$) traps was there a significant difference in plum curculio response to bait treatments (Fig. 5).

Late Season

In this period, each trap type captured similar numbers of adults (df = 1, 64; F = 0.15; $P > 0.05$) (mean ± SE, 0.55 ± 0.10 and 0.50 ± 0.12 for panel and pyramid traps,

respectively). Panel traps baited with benzaldehyde plus pheromone captured significantly more adults than any other panel traps (df = 7, 64; F = 5.56; $P < 0.0001$), among which there were no significant differences. Pyramid traps baited with benzaldehyde plus pheromone captured significantly more adults than any other pyramid traps. Pyramid traps baited with limonene plus pheromone captured significantly more plum curculios than pyramid traps baited with either decyl aldehyde plus pheromone or *E*-2-hexenal plus pheromone (df = 7, 64; F = 6.13; $P < 0.001$) (Fig. 6).

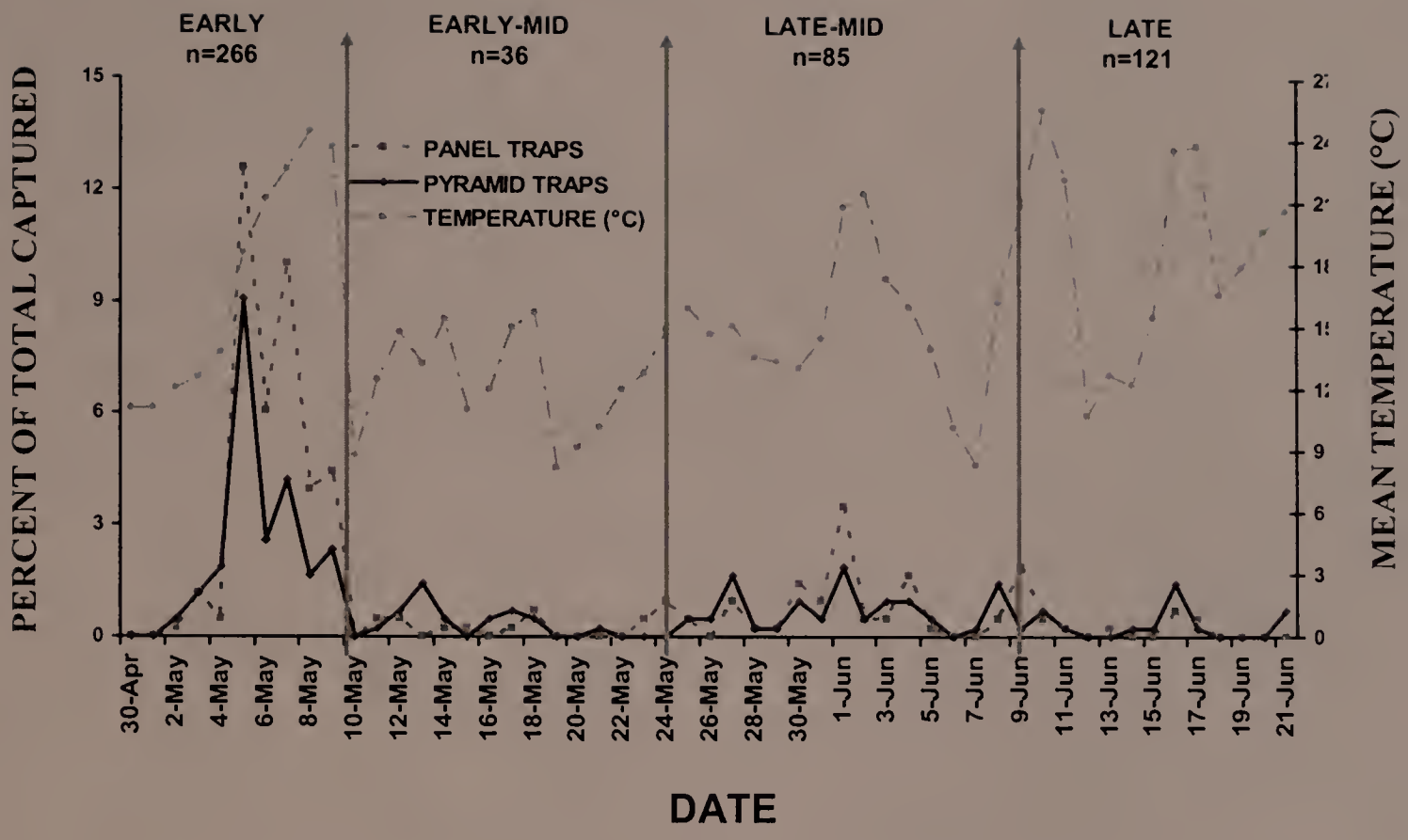

Figure 3. Percentage of seasonal total number of adult plum curculios captured per day, according to trap type. Mean daily temperature (°C) at study site is also shown.

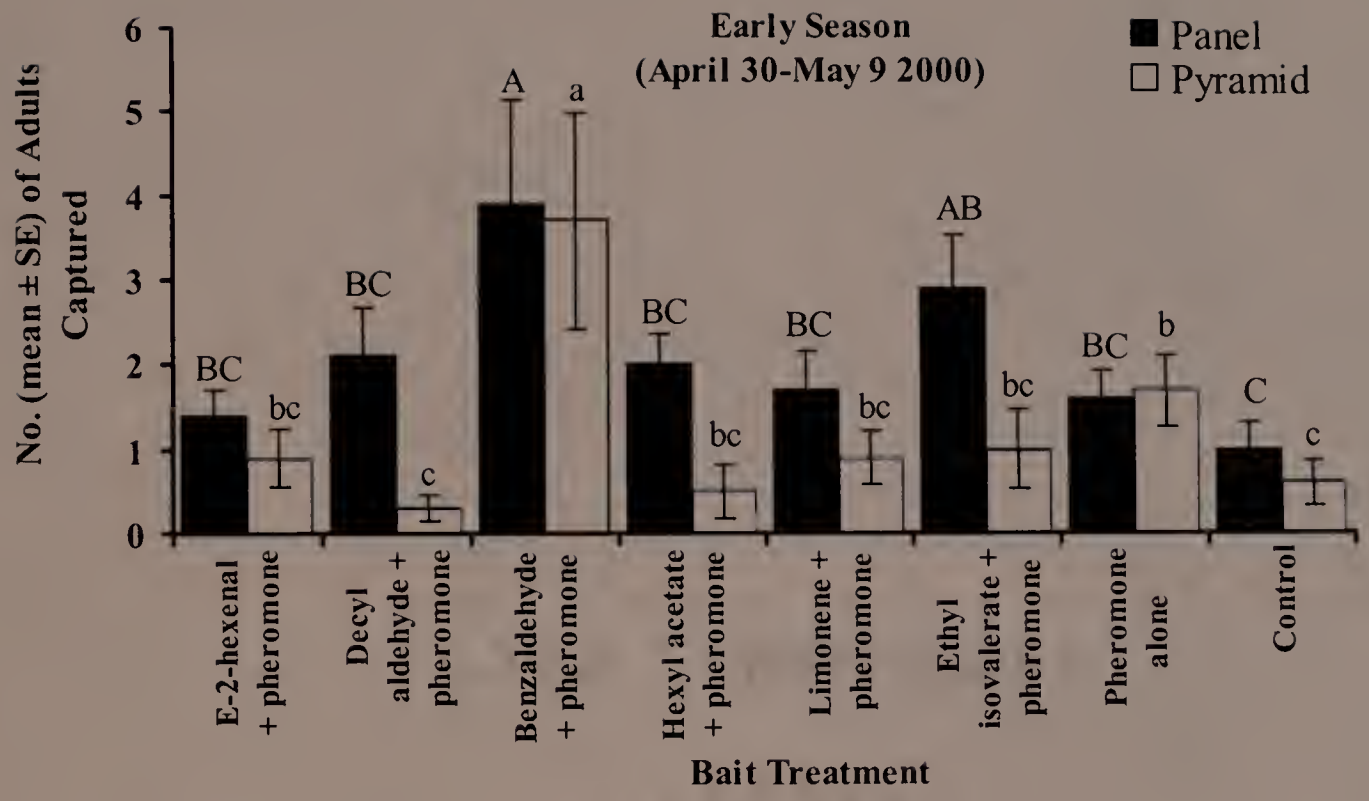

Figure 4. Plum curculio captures (mean ± SE) according to trap type and bait treatment in early season (30 April-9 May 2000). Bars superscribed by the same letter are not significantly different from one another ($\alpha = 0.05$; LSD test).

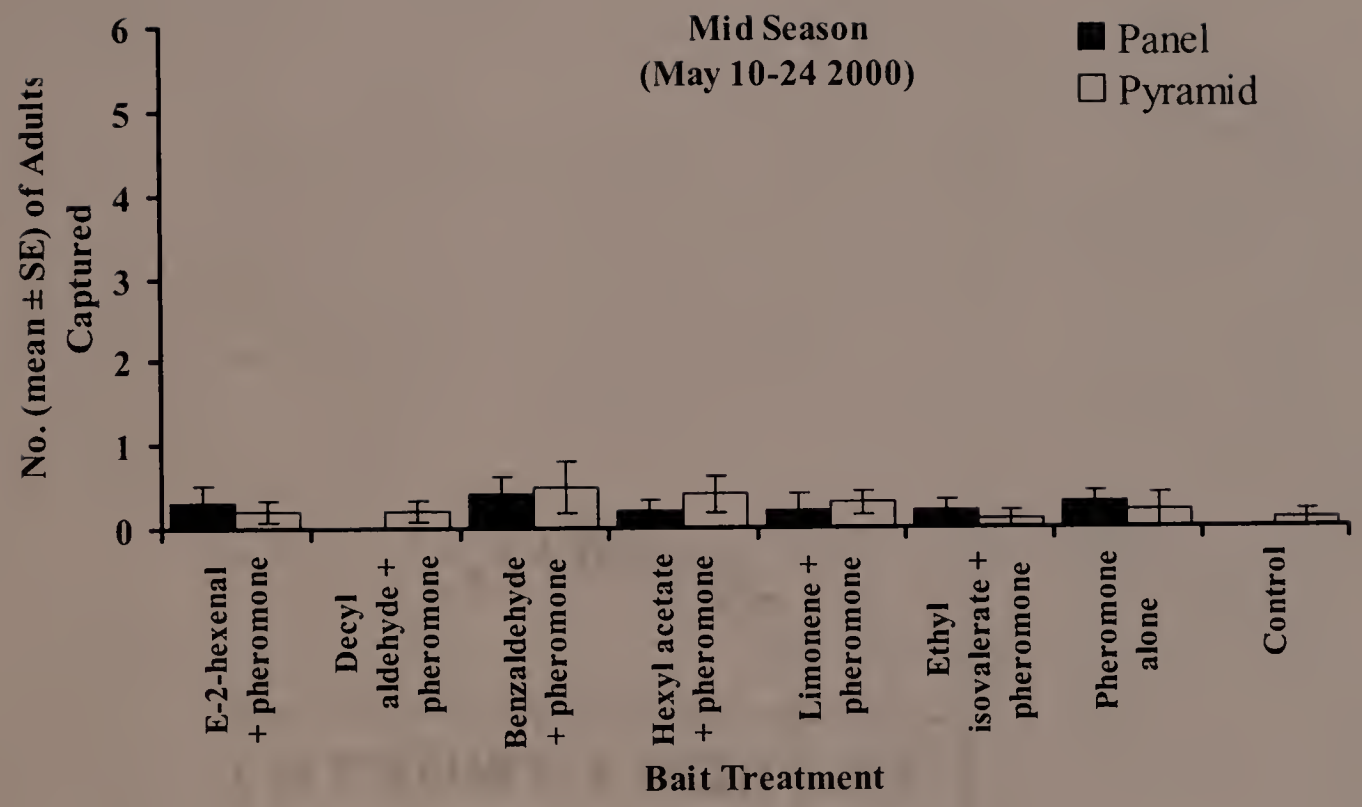

Figure 5. Plum curculio captures (mean ± SE) according to trap type and bait treatment in mid season (10-24 May 2000). No significantly differences were found among bait treatments for either trap type ($\alpha = 0.05$; LSD test).

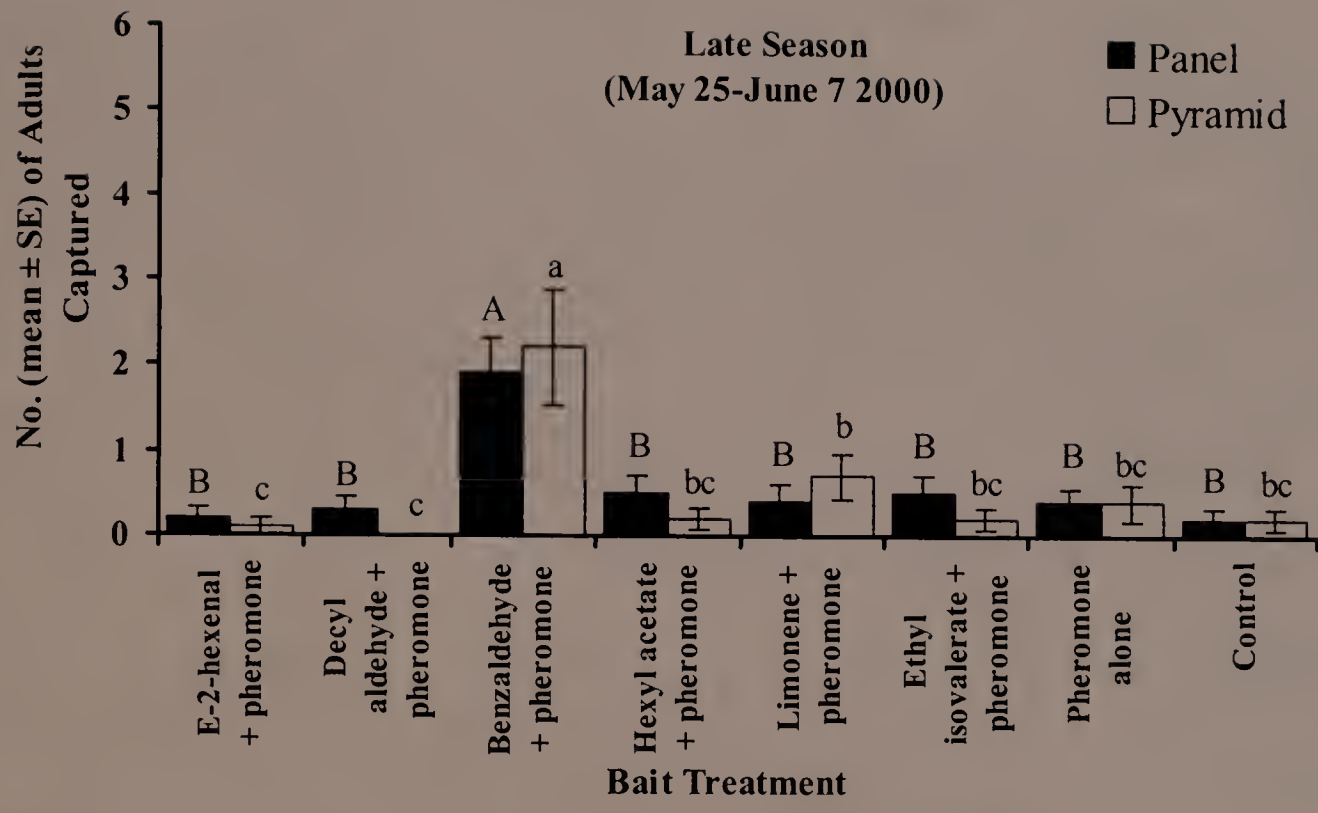

Figure 6. Plum curculio captures (mean ± SE) according to trap type and bait treatment in late season (25 May-7 June 2000). Bars superscribed by the same letter are not significantly different from one another ($\alpha = 0.05$; LSD test).

Trap Captures According to Time of Day

Panel traps captured nearly twice as many adults from 1430 to 1930 hours as from 1930 to 0730 hours, and nearly 4 times as many as from 0730 to 1430 hours (df = 2, 27; F = 6.05; $P = 0.007$) (Fig. 10). In contrast, pyramid traps captured numerically fewer adults from 1430 to 1930 hours than from 1930 to 0730 hours, with very few captured from 0730 to 1430 hours (df = 2, 27; F = 5.28; $P = 0.01$) (Fig. 7. Mean, maximum and minimum temperature values registered for each time period are also depicted in Fig. 7.

Influence of Weather Factors

Results of stepwise multiple regression analyses (Table 2) indicated that across all three seasonal time periods, responses of both males and females to panel traps were positively and significantly correlated with temperature for bait treatments of benzaldehyde plus pheromone as well as pheromone alone. For pyramid traps baited with benzaldehyde plus pheromone, male and female responses were independent of temperature. In contrast, for pyramid traps baited with pheromone alone, both male and female captures were positively and significantly correlated with temperature, and female captures were positively and significantly associated with wind speed. In all cases, temperature exerted a stronger influence (R^2 values were greater) on captures by panel traps than by pyramid traps (Table 2). In no case was relative humidity, rainfall, or barometric pressure significantly associated with captures by either type of trap.

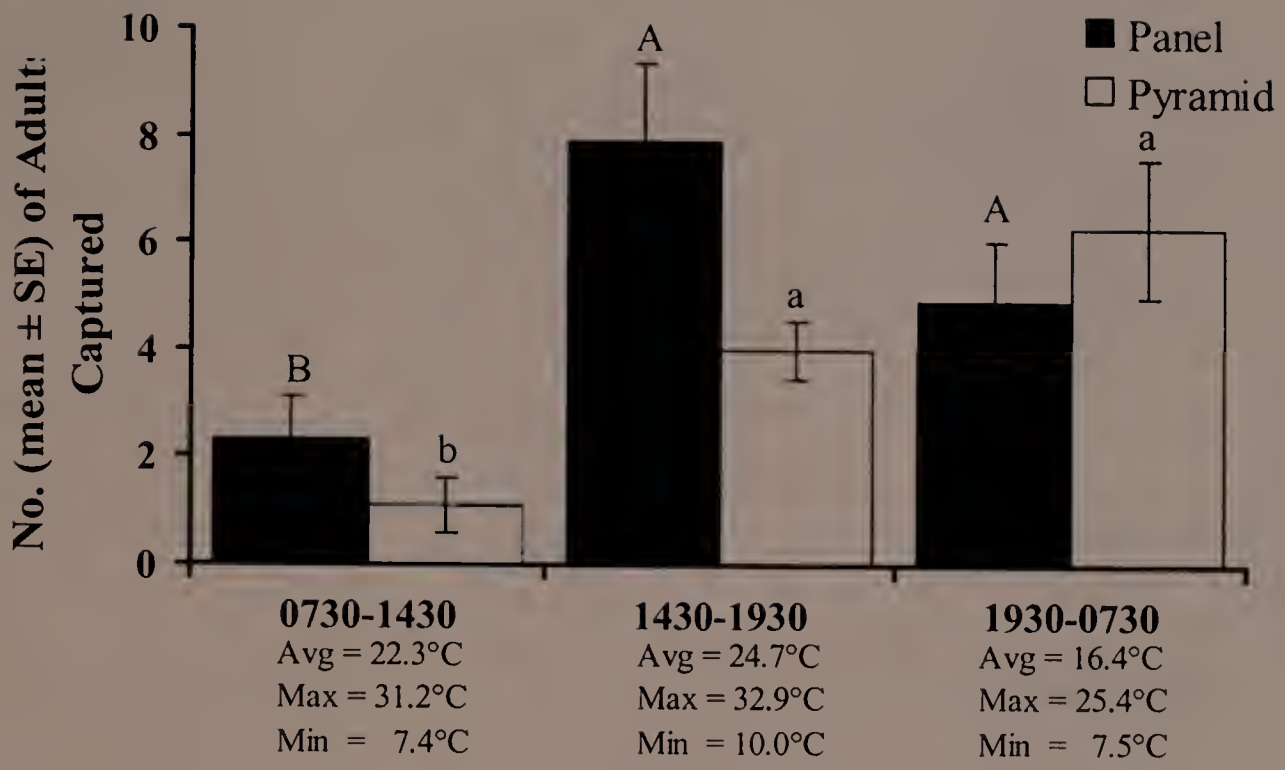

Figure 7. Plum curculio captures (mean ± SE) by panel and pyramid traps according to time of day. Mean (Avg.), maximum (Max), and minimum (Min) temperature values registered for each time period are presented.

Discussion

My results indicate that early in the season (from tight cluster to bloom: 30 April-9 May), when plum curculios were presumably immigrating into the apple orchard, panel traps placed close to woods and baited with benzaldehyde plus pheromone or ethyl isovalerate plus pheromone were about 4- and 3-fold, respectively, more attractive to plum curculios than were unbaited panel traps. During this period, pyramid traps baited with benzaldehyde plus pheromone or pheromone alone were ca. 6- and 3-fold, respectively, more attractive to plum curculios than were unbaited pyramid traps. There was little response to traps of any type during mid-season (i.e., during petal-fall: 10-24 May). Throughout late season (i.e., fruit-set: 25 May-7 June), plum curculio response to panel and pyramid traps baited with benzaldehyde plus pheromone was ca. 6- and 11-fold, respectively, greater than to unbaited traps of each type.

Initial captures of immigrating plum curculios (three adults) by traps occurred on 2 May, during the tight-cluster stage, and the peak of immigration was observed on 5 May (during full bloom), a day on which 93 adults (24% of season-long total) were captured by traps. Coincidently, the earliest reported adult appearance in host apple trees after overwintering has been recorded as occurring at tight cluster (Lafleur and Hill 1987).

Of particular interest are the substantial early-season captures of plum curculios by panel and pyramid traps baited with benzaldehyde plus pheromone, and to a lesser extent by panel traps baited with ethyl isovalerate plus pheromone. Release rate seems to play an important role in plum curculio response to benzaldehyde and ethyl isovalerate.

Here, these compounds were tested at a moderate release rate (~10 mg/d) in combination with pheromone. Benzaldehyde when alone was considered by Prokopy et al. (2001) as only mildly attractive at a low release rate (1.3 mg/d) and by Leskey et al. (2001) as insignificantly attractive when at a high release rate (48 mg/d) under field conditions. Prokopy et al. (2000) found no significant differences in plum curculio captures by pyramid, cylinder, or Circle traps baited with a combination of ethyl isovalerate, limonene (both at a high release rate), and grandisoic acid when compared with unbaited traps placed in association with apple trees in unsprayed or commercial orchards. However, in their study, panel traps baited with this same odor combination captured ca. 10 times more adults than unbaited traps when positioned adjacent to woods, suggesting that type or position of trap and/or release rate of odor may have affected degree of plum curculio response to traps. My results are encouraging in showing that both benzaldehyde plus pheromone and ethyl isovalerate plus pheromone may have promise as lures for attracting immigrating plum curculios in early spring.

Lack of attractiveness of benzaldehyde plus pheromone over the 15 d of the mid-season period could have been due, in part, to impact of low temperatures on level of adult activity but also in part to loss of activity of pheromone and altered activity of benzaldehyde. I subsequently detected polymerization of ca. 10% of the benzaldehyde contained in vials into a secondary (yellow, crystalline solid) unidentified chemical, which may have diminished or masked the attractiveness of benzaldehyde. Such polymerization may have begun during the warm days that characterized the early-season period. After replacing vials containing benzaldehyde and replacing pheromone dispensers (at 24 d following initial deployment), a strong positive response to this

combination was again observed (during late season). Thus, a challenge for the future is to find an improved approach for dispensing benzaldehyde to ensure stability and longevity at a desired release rate.

In mid- and late-season, ethyl isovalerate plus pheromone was not significantly attractive to plum curculios, as was the case in early-season when in association with pyramid traps. Thus, a possible progressive loss of attractiveness of ethyl isovalerate or, alternatively, a season-specific response of plum curculios to this chemical may have occurred. Vials containing this chemical were not replaced because I had estimated that the activity would endure for ~97 d, but it is possible that the properties of ethyl isovalerate may have changed over time.

Some studies (Quaintance and Jenne 1912, Smith and Flessel 1968) have shown that temperature exerts a strong effect on emergence patterns of plum curculios after overwintering. Other studies (Dixon et al. 1999, Prokopy et al. 1999b) have attempted to correlate the influence of weather on plum curculio captures by traps. In our study, periodic peaks of adult captures by traps seemed to coincide with rises in temperature suggesting that, in early spring, temperature may be the most important factor favoring adult dispersal and orchard colonization, as suggested by Smith and Flessel (1968) for plum curculio and by Duan et al. (1996) and Toepfer et al. (1999) for the apple blossom weevil. In fact, when captures of adults by traps were studied in association with the five weather factors evaluated here (temperature, relative humidity, barometric pressure, rainfall, and wind speed), I found that temperature was positively and significantly correlated with plum curculio captures by both trap types, except for male and female

captures by pyramid traps baited with benzaldehyde plus pheromone, which were weather-independent.

The stronger positive correlation found between temperature and adult captures by panel traps baited with benzaldehyde plus pheromone or pheromone alone than between temperature and captures by pyramid traps baited with these lures may be associated with the mode of action of each trap type. Dixon et al. (1999) found a strong positive correlation between adult captures by unbaited panel traps and ambient temperature, but no weather factors significantly influenced captures of adults by unbaited pyramid traps, thus this type of trap may be less weather-dependent. Nevertheless, it has been shown that adult captures by pyramid traps on cool days (< 19°C) may be greater than during warm days (> 20°C) because at lower temperature, adults reentering host trees do so almost exclusively by walking toward and onto tree trunk silhouettes (Prokopy et al. 1999b).

An oviposition model that relates temperature (cumulative heat units) to cumulative fruit injury for scheduling insecticide applications against plum curculio was developed recently by Reissig et al. (1998). Based on my results, I believe that addition to this model of a profile of captures of immigrating adults by traps would be beneficial, particularly if adult capture patterns could be correlated adequately with fruit injury by plum curculio in commercial orchards. The latter is a critical aspect that has been investigated by Prokopy et al. (1999a, 2000) but without substantial success as yet.

In summary, my findings indicate that both panel and pyramid traps baited with benzaldehyde plus pheromone were useful in detecting early plum curculio arrivals (at tight cluster stage) in an apple orchard, allowing us to also gain information on the peak

(at full-bloom stage) and end of immigration (at fruit set). I conclude that placement of panel and/or pyramid traps baited with benzaldehyde plus pheromone at margins of plum curculio overwintering sites (particularly near areas of orchards where plum curculio injury is known from previous experience to be most likely) may be valuable for identifying the beginning, peak, and possibly also the end of plum curculio immigration. Such knowledge can aid in accurate timing of insecticide application. Further research on combinations and release rates of odor attractants and trap placement could improve the performance of traps for this purpose.

Table 1. Number of dispensers placed per trap, release rate from dispensers, and expected duration of each compound used in association with a trap as determined under laboratory conditions. Vials containing benzaldehyde and limonene and all pheromone dispensers were replaced after 24 d to maintain consistent release rate

Compound	No. Vials/Trap	Release Rate, (mg/d)[a]	Expected Duration (days)
E-2-hexenal	5	9.65	75
Decyl aldehyde	4	9.79	136
Benzaldehyde	4	10.11	165
Hexyl acetate	3	10.71	98
Limonene	2	9.50	35
Ethyl isovalerate	3	10.72	97
Grandisoic acid	1 dispenser	1.00	24

[a]Release rates of each synthetic fruit volatile represent mean daily release from experimental dispensers used. These data were based on an empirical study performed by us under laboratory conditions in which loss of mass from each vial of each chemical was recorded daily for 30 d by using an analytical scale (± 0.01 mg).

Table 2. Relationships between weather conditions (temperature, relative humidity, rainfall, barometric pressure, and wind speed) and plum curculio captures by traps according to sex, trap type, and bait across the entire season (30 April-7 June 2000). BEN+GA= benzaldehyde plus grandisoic acid; GA= grandisoic acid.

Sex	Trap Type	Bait	Weather Factor	df	t^a	P	R^2
Male	Panel	BEN+GA	Temperature	1, 36	2.9	0.01	0.35
Male	Pyramid	BEN+GA	None				
Male	Panel	GA	Temperature	1, 36	3.3	0.003	0.54
Male	Pyramid	GA	Temperature	1, 36	2.3	0.03	0.25
Female	Panel	BEN+GA	Temperature	1, 36	3.5	0.003	0.44
Female	Pyramid	BEN+GA	None				
Female	Panel	GA	Temperature	1,36	3.4	0.03	0.42
Female	Pyramid	GA	Temperature	1, 36	3.1	0.004	
			Wind speed	2, 35	2.2	0.04	0.32^b

[a]All values presented indicate positive correlations

[b]R^2 combined at step 2 of regression analysis

CHAPTER 3

FIELD EVALUATION OF PLANT ODOR AND PHEROMONAL COMBINATIONS FOR ATTRACTING PLUM CURCULIOS

Abstract

The attractiveness of different synthetic host odors and a synthetic aggregation pheromone (grandisoic acid) to overwintered adult plum curculios, *Conotrachelus nenuphar* (Herbst) (Coleoptera: Curculionidae), was examined using two types of traps (sticky panels and black pyramids) placed in border areas surrounding an unsprayed section of an apple orchard in Massachusetts. In 2001, I evaluated the response of plum curculios to three synthetic fruit volatiles (benzaldehyde, ethyl isovalerate, and limonene) assessed alone and in combination with grandisoic acid, as well as response to grandisoic acid alone and a no-odor (control) treatment. Benzaldehyde was the only host volatile that for both trap types synergized the response of plum curculios to grandisoic acid. For both trap types, grandisoic acid was just as attractive to plum curculios when alone as when in combination with either ethyl isovalerate or limonene. In 2002, four release rates of benzaldehyde (0, 2.5, 10, and 40 mg/day) and two release rates of grandisoic acid (1 and 2 mg/day) were evaluated for attractiveness to plum curculios using panel and pyramid traps. For panel traps, an increase in amount of grandisoic acid released (from 1 to 2 mg/day) was associated with a 35% increase in captures. However, plum curculio captures by pyramid traps were similar regardless of the amount of grandisoic acid

released. For panel traps, 10 and 40 mg/day of benzaldehyde were the most attractive release rates regardless of the amount of grandisoic acid released. For pyramid traps baited with grandisoic acid, plum curculio captures were enhanced by the presence of benzaldehyde, regardless of release rate. In 2003, when in association with grandisoic acid at 1 mg/day of release, benzaldehyde at 80 mg/day of release did not enhance plum curculio captures by panel traps relative to lower release rates of benzaldehyde evaluated. Pyramid traps releasing grandisoic acid at 1 mg/day performed best when baited with benzaldehyde at 10 mg/day of release; a release rate of 80 m/day of benzaldehyde decreased significantly the attractiveness of the binary combination of benzaldehyde + grandisoic acid. Combined results suggest that benzaldehyde at 10 mg/day of release in association with grandisoic acid at 1 mg/day of release constitutes a powerful lure that may greatly improve the effectiveness of monitoring traps for plum curculios.

Introduction

Aggregation via behavior-modifying chemicals occurs in a wide range of insects, but predominantly in the Coleoptera (Borden, 1984; Landolt, 1997). This behavioral feature has allowed researchers and integrated pest management practitioners to develop and implement semiochemical-based trapping of adults of a number of beetle species that are pests in both agroecosystems and forests (Borden, 1993). For instance, the discovery that aggregation pheromones and natural host-produced volatiles mediate host selection behavior in weevils has led to the development of viable monitoring and mass trapping systems for several species of beetles. Thus, in bark beetles, much attractiveness is

achieved when synthetic aggregation pheromones are used in association with live or dead host trees (Borden 1984; Byers 1989; Landolt and Phillips 1997). Analogously, for species of weevils in which mass trapping is used as a control tactic, such as *Rhynchophorus palmarum* L. (Alpizar et al., 2002; Oehlschlager et al., 2002a), *R. ferrugineus* (Olivier) (Hallett et al., 1999), and *Metamasius hemipterus* L. (Ramirez-Lucas et al., 1996; Alpizar et al., 2002; Oehlschlager et al., 2002b), the most powerful attractants involve use of aggregation pheromones, synthetic host plant volatiles and host plant material. Recent investigations by Phillips et al., (1993), Gries et al., (1994), Perez et al., (1994), Weissling et al., (1994), Ramirez-Lucas et al., (1996), Perez et al., (1997), and Hallet et al., (1999) have shown that either natural host tissue or synthetic host plant volatiles are able to synergistically enhance weevil response to aggregation pheromone.

The plum curculio, *Conotrachelus nenuphar* (Herbst.), is a key pest of pome and stone fruit in eastern and central North America (Racette et al., 1992). In the fall, most adults migrate from apple orchards to the edges of adjacent forested areas, where they hibernate (Lafleur et al., 1987). In spring, overwintered adults immigrate into apple orchards, where they cause injury to apple buds and developing fruit as a result of adult feeding and oviposition activity, the latter commencing shortly after petal fall (Lafleur and Hill, 1987; Chouinard et al., 1993). Despite its status as a key pest, no truly effective traps have yet been developed for use in the northeastern North America for tracking the seasonal course of plum curculio immigration in a way that allows growers to implement management tactics against plum curculio based on information from traps (reviewed in Prokopy et al., 2003). In the southern USA, baited pyramid traps have been proposed as a useful sampling tactic for plum curculio in peach orchards, where they may aid growers

in determining need and timing of insecticide application against plum curculio based on trap captures and thus (Johnson et al. 2002).

Plum curculios use chemical cues to locate potential hosts (Butkewich and Prokopy, 1993). Based upon screening of more than 30 individual compounds, recent investigations have revealed that some volatiles such as benzaldehyde, ethyl isovalerate, and limonene, released from developing host fruit, are attractive to plum curculios (Leskey and Prokopy, 2000; Leskey et al., 2001; Prokopy et al., 2001). In particular, it has been shown that benzaldehyde, when used in combination with the plum curculio aggregation pheromone grandisoic acid (Eller and Bartelt, 1996), is a highly attractive lure for plum curculios (chapter 2; Prokopy et al., 2003). Relative to other combinations of synthetic fruit odor and grandisoic acid, this odor combination enhanced significantly the performance of both clear sticky Plexiglas panels and black pyramid traps (two promising plum curculio monitoring devices) deployed near woods adjacent to apple trees (chapter 2).

Given the promise of these compounds as synthetic lures for plum curculio, it becomes important to determine potential additive or synergistic relationships between synthetic fruit odors and grandisoic acid, as well as to determine the amount of fruit odor and grandisoic acid needed to improve the performance of panel and pyramid traps when deployed in apple orchards. Here, in a 2001 study, I aimed at evaluating the attractiveness of three different synthetic host volatiles for plum curculio (benzaldehyde, ethyl isovalerate, and limonene) alone and in combination with grandisoic acid. By doing so, I sought to determine the extent to which each synthetic host volatile enhanced plum curculio response to grandisoic acid. In a 2002 experiment, I evaluated plum curculio

response to four different release rates of benzaldehyde (the most attractive host odor found in 2001) and two release rates of grandisoic acid to determine the amount of each odor needed to maximize the performance of monitoring devices. In a 2003 study, my main objective was to determine the maximum release rate at which benzaldehyde in combination with grandisoic acid was attractive to plum curculios, relative to release rates of benzaldehyde evaluated in 2002.

Materials and Methods

Studies were carried out in 2001, 2002, and 2003 in an unsprayed section of apple trees at the University of Massachusetts Cold Spring Orchard in Belchertown, MA. The perimeter of the unsprayed section was about 500 m long and bordered almost entirely by woods.

Traps

For each of the three years, two trap types were used: (a) clear Plexiglas panels (60 x 60 cm, with the side facing the woods coated with Tangletrap), intended to capture plum curculios in flight (particularly during warm days), and (b) black pyramid traps (61 cm wide at base x 122 cm tall, modified after Tedders and Wood, 1994), intended primarily to capture crawling plum curculios, mostly during cool days or at night. Trap design is described in detail in chapter 2.

Synthetic Host Volatiles in Association with Grandisoic Acid

The host volatiles tested in 2001 were benzaldehyde (99.5%), ethyl isovalerate (98%), and limonene ([*R*]-[+]-limonene, 97%), all obtained from Sigma-Aldrich Chemical Co. (Milwaukee, WI). These three synthetic odors had previously been shown to be attractive to plum curculios (Leskey et al., 2001; Prokopy et al., 2001), particularly when in association with grandisoic acid (chapter 1). Three different groups of odor treatments were arranged, each involving a single host volatile alone, grandisoic acid alone, a combination of host volatile and grandisoic acid, and a no-odor (control) treatment. There were four replicates for each trap type and group of treatments arranged in a randomized complete block design.

Benzaldehyde was released from 1-ml low-density white polyethylene vials (Wheaton, Millville NJ) to prevent polymerization of this chemical by UV light, as may have occurred in a previous field study (chapter 2) that involved use of clear polyethylene vials. Because this shortcoming was not found for ethyl isovalerate and limonene, these two chemicals were evaluated using 400µl high-density clear polyethylene vials (VWR Scientific Products, Boston, MA). Two 400µl vials containing limonene (in a 50:50 ratio of limonene and mineral oil to slow the release rate) and three vials containing ethyl isovalerate (each filled with 400µl of ethyl isovalerate) were needed per trap to accomplish a release rate of ~10 mg/day. A white vial filled with 1 ml of benzaldehyde released ~2.5 mg/day. Four such vials per trap were used, giving a release rate of ~10 mg/day/trap. Each pheromone dispenser (ChemTica International, S.A., San Jose, Costa Rica) released ~1 mg/day of grandisoic acid. Release rates of grandisoic acid and

synthetic fruit volatiles were determined outdoors by estimating the daily mean weight loss of each chemical using an analytical balance (±0.01mg).

Vials containing host volatiles were attached to the lower edge of panels using steel binder clips. For pyramid traps, vials were placed inside a screen funnel (boll weevil trap top; Gempler's, Mt. Horeb, WI) that capped each trap. A single grandisoic acid-releasing dispenser was attached to the upper edge of a Plexiglas panel using a binder clip, or was positioned inside the boll weevil trap top. Vials containing benzaldehyde were replaced approximately every two weeks during May and June to ensure consistent release. Vials containing ethyl isovalerate and limonene, along with grandisoic acid dispensers, were replaced twice, once in mid-May and once in mid-June.

Evaluation of Different Release Rates of Benzaldehyde and Grandisoic Acid

In 2002, eight different odor treatments, dictated by a combination of four different release rates of benzaldehyde (0, 2.5, 10, and 40 mg/day) and two release rates of grandisoic acid (1 and 2 mg/day), were evaluated for attractiveness to plum curculios. The low release rate of benzaldehyde (~2.5 mg/day) was achieved by using a 1-ml low-density white polyethylene vial filled with 1 ml of benzaldehyde per trap, as in our 2001 study. The medium release rate (~10 mg/day) was achieved by using one 15-ml low-density white polyethylene vial, filled with 15 ml of benzaldehyde, per trap. The high release rate (~40 mg/day) was achieved by using four such vials per trap. Because each pheromone dispenser released on average ~1 mg of grandisoic acid per day, the high release rate of grandisoic acid (2 mg/day) was achieved by using two grandisoic acid

dispensers per trap. There were six replicates for each trap type and odor combination, following a randomized complete block design.

Each vial containing benzaldehyde (either a 1-ml or a 15-ml vial, depending on the treatment) was hung by its neck from a wire and placed inside an inverted green 266-ml plastic cup (CVS, Woonsocket, RI) to provide additional protection for this chemical against rainfall and polymerization by UV light. Cups holding benzaldehyde-dispensing vials were attached to the bottom edge of panels using wire and steel binder clips. Cups were hung from the end of a wooden pole (buried in the ground at a 45° angle) in such a way that bases of cups were ~10 cm above pyramid trap tops. Either one or four cups were attached to each pole, depending on the treatment. Grandisoic acid dispensers were attached to the upper edge of panels using binder clips, or were placed inside the inverted screen funnel capping pyramid traps. Benzaldehyde-releasing vials and grandisoic acid dispensers were replaced once, four weeks after trap deployment.

2003 Evaluation of Benzaldehyde

In 2003, I evaluated benzaldehyde at 0, 10, 40, and 80 mg/day of release, in association with grandisoic acid at 1 mg/day of release, with the aim of determining if a further increase of benzaldehyde (relative to the amounts of benzaldehyde evaluated in 2002) was associated with an increase in plum curculio captures by traps. The first three treatments (i.e., benzaldehyde at 0, 10, and 40 mg/day of release) evaluated in 2003 were considered as baseline. Benzaldehyde was dispensed as in the 2002 study with the difference that (1) benzaldehyde was mixed in a 9:1 ratio with 1, 2, 4-Trichlorobenzene

(99+%) (Sigma-Aldrich Chemical Co., Milwaukee, WI) as a stabilizing chemical (S.E., Wright, personal communication), and (2) I used vials containing 7.5 ml of the mixture, releasing on average ~10 mg of benzaldehyde per day. Vials releasing benzaldehyde were not replaced throughout the study. Grandisoic acid dispensers were replaced once, five weeks after trap deployment. There were four replicates for each trap type and odor treatment, arranged in a randomized complete block design.

Trap Deployment

In 2001 and 2002, traps were deployed in pairs (one of each type spaced 1 m apart) along the periphery of the orchard, in close proximity (~1 m) to woods. Each pair of traps was baited with the same odor treatment and spaced 10 m from other trap pairs on either side. In 2003, panel and pyramid traps were spaced 10 m apart to ensure that adult response was solely to intended release rate and not to double the intended release rate that in 2001 and 2002 may have occurred as a consequence of a panel and a pyramid trap having been deployed in close proximity (i.e., 1 m apart).

Trap deployment and baiting took place during the tight cluster stage of bud development (on April 30, 16, and 28, in 2001, 2002, and 2003, respectively). Traps were inspected for plum curculio captures on a daily basis, although results for each year show plum curculio captures by panel or pyramid traps across the entire period of immigration. To minimize the influence of trap location on adult captures I rotated, within each replicate, the bait-containing portion of each trap by one position. This procedure was performed clockwise 12 times in 2001, and eight times in 2002 and 2003.

Statistical Analysis

For all analyses, the response variable was the number of adult plum curculios captured by traps of each type across the total number of days comprising the period of plum curculio immigration (62 days in 2001, 82 days in 2002, and 77 days in 2003). Results present total plum curculio captures (males and females combined), as preliminary analyses showed that for all three years, males and females responded similarly to the odor treatments and trap types. Data were subjected to analysis in a non-transformed fashion as the homogeneity-of-variances assumption was met in all cases. For all ANOVAS performed, block was used as random factor. For each analysis performed, Fisher-protected LSD separations of treatment means were performed when appropriate. All statistical analyses were performed using Statistica® (StatSoft, 2001).

In 2001, in addition to ANOVA, I performed comparisons of ratios of interaction (ROIs) (Hammack, 1996) to examine type of interaction (inhibitory, additive, or synergistic) among single vs. 2-component odor treatments. In our case, a ROI= [(A + grandisoic acid) + control]/[(A) + (grandisoic acid)], where (A) represents plum curculio captures by traps baited with a particular fruit volatile alone, (grandisoic acid) is the capture of plum curculios by traps baited with grandisoic acid alone, (A+grandisoic acid) denotes plum curculio captures by traps baited with a particular fruit volatile in association with grandisoic acid, and control represents plum curculio captures by unbaited traps. ROI values significantly inferior, equal, or superior to 1 offer evidence for inhibitory, additive (= neutral effect), or synergistic interactions, respectively, between chemicals (Hammack, 1996). Each ROI value obtained corresponded to one of the four

replicates performed for each odor group and trap type. Subsequently, a two-tailed Student's t test using such ROI values was performed to test the null hypothesis of ROI= 1 for each fruit odor evaluated (benzaldehyde, ethyl isovalerate and limonene). Further, to verify the possible occurrence of synergism in the particular case of benzaldehyde, after performing the ANOVA I compared plum curculio response to traps baited with benzaldehyde + grandisoic acid against the combined response of plum curculios to traps baited with either benzaldehyde or grandisoic acid alone using linear orthogonal contrast analysis, which allows for specific comparisons of means of the treatments of interest by use of sets of contrast weights (StatSoft, 2001).

Results

Attractiveness of Synthetic Host Volatiles in Association with Grandisoic Acid

In 2001, benzaldehyde in combination with grandisoic acid was the most attractive odor combination for plum curculios for both panel (ANOVA df= 3, 9; F= 24.39; $P< 0.001$) and pyramid (ANOVA df= 3, 9; F= 5.36; $P= 0.02$) traps. Figure 11A shows that panel traps baited with benzaldehyde+grandisoic acid captured significantly more plum curculios than did benzaldehyde-baited, grandisoic acid-baited or unbaited panel traps. Plum curculio response to grandisoic acid alone was significantly greater than response to traps baited with benzaldehyde alone or to unbaited traps. Likewise, as shown in figure 8B, pyramid traps baited with benzaldehyde+ grandisoic acid captured significantly more plum curculios than did benzaldehyde-baited, grandisoic acid-baited or

unbaited pyramid traps. When baited with benzaldehyde alone or grandisoic acid alone, pyramid traps captured significantly more plum curculios than did unbaited pyramid traps. Comparisons of ROI values (Table 3), as well as contrast analysis, provided evidence that for both trap types benzaldehyde synergistically increased the response of plum curculios to grandisoic acid.

For both trap types, the addition of ethyl isovalerate did not increase the attractiveness of grandisoic acid to plum curculios (Fig. 8A and B). For both trap types, significantly more plum curculios were captured by traps baited with grandisoic acid alone than by traps baited with ethyl isovalerate alone or by control traps (ANOVA df= 3, 9; F= 10.57; $P< 0.01$ and df= 3, 9; F= 12.72; $P< 0.001$ for panel and pyramid traps, respectively). In no case did ethyl isovalerate alone significantly enhance adult response above that to control traps. Comparisons of ROI values provided evidence that ethyl isovalerate interacted with grandisoic acid in an additive manner (Table 3).

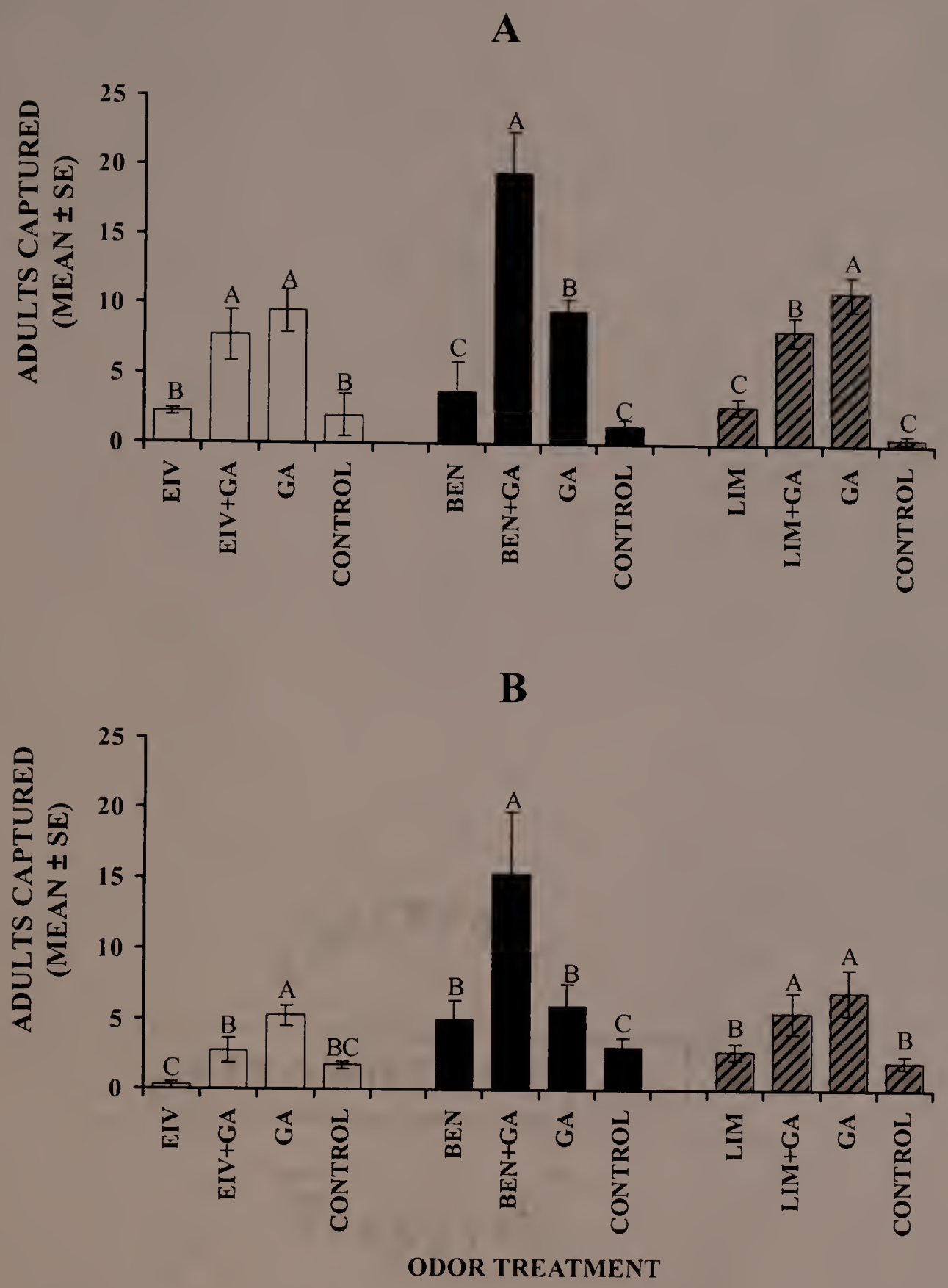

Figure 8. Plum curculio captures (mean ± SE) by A) panel traps and B) pyramid traps baited with three different synthetic fruit volatiles alone or in combination with grandisoic acid (GA) in 2001. EIV= ethyl isovalerate, BEN= benzaldehyde, LIM= limonene. For each graph, bars in each group not superscribed by the same letter are significantly different from each other according to ANOVA ($P< 0.05$) and LSD tests.

For both trap types, the presence of limonene had no positive effect on the attractiveness of grandisoic acid to plum curculios. Relative to limonene-baited traps or control traps, panel and pyramid traps baited with grandisoic acid alone or with grandisoic acid +limonene captured significantly more plum curculios (ANOVA df= 3, 9; F= 26.57; $P< 0.001$ and df= 3, 9; F= 4.14; $P= 0.04$ for panel and pyramid traps, respectively). For panel traps, however, grandisoic acid attracted significantly more plum curculios when alone than when in combination with limonene, whereas for pyramid traps both treatments attracted similar numbers of plum curculios. Captures by traps of either type baited with limonene alone did not differ from captures by unbaited traps. For panel traps, a ROI value of 0.66 indicated an inhibitory effect when limonene (at the release rate evaluated) was added to grandisoic acid. For pyramid traps, a ROI value of 0.90 denoted that the interaction between limonene and grandisoic acid was additive (Table 3).

Evaluation of Different Release Rates of Benzaldehyde and Grandisoic Acid

In 2002, I found that panel traps baited with two grandisoic acid dispensers, releasing 2 mg/day of grandisoic acid on average, captured about 35% more plum curculios than did panel traps baited with only one grandisoic acid dispenser (releasing, on average, 1 mg/day of grandisoic acid) (ANOVA df= 1, 5; F= 6.95; $P= 0.04$). In contrast, pyramid traps captured similar numbers of plum curculios regardless of the amount of grandisoic acid released (ANOVA df= 1, 5; F= 2.31; $P= 0.18$) (Fig. 9).

Figure 10A reveals that, for panel traps baited with only one grandisoic acid dispenser (1 mg/day of release), benzaldehyde at 40 mg/day of release attracted the most plum curculios, followed by benzaldehyde at 10 mg/day of release (ANOVA df= 3, 15; F= 4.41; *P*= 0.02). For panel traps baited with two grandisoic acid dispensers (2 mg/day of release), differences among benzaldehyde treatments were only numerical and not statistically significant at *P*< 0.05 (ANOVA df= 3, 15; F= 2.77; *P*= 0.08).

Figure 10B shows that, for pyramid traps baited with the low release rate of grandisoic acid (1 mg/day), the mere addition of benzaldehyde (regardless of amount released) enhanced numerically but not significantly plum curculio captures relative to traps baited with grandisoic acid alone (ANOVA df= 3, 15; F= 2.16; *P*= 0.13). For pyramid traps baited with the high release rate of grandisoic acid (2 mg/day), I found that addition of benzaldehyde at either 2.5 or 10 mg/day (but not 40 mg/day) did significantly enhance plum curculio response relative to grandisoic acid alone (ANOVA df= 3, 15; F= 4.68; *P*= 0.02).

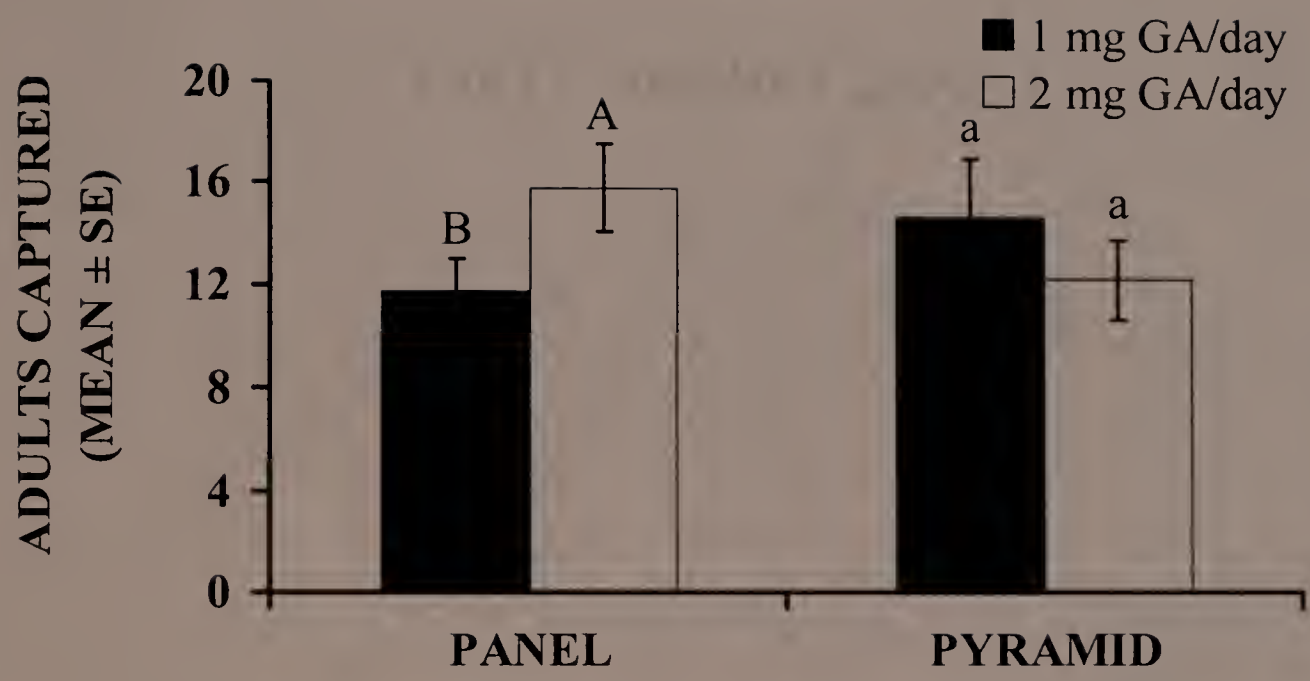

Figure 9. Plum curculio captures (mean ± SE) by panel and pyramid traps as a function of amounts of grandisoic acid released per day in 2002. Data were pooled across all amounts of benzaldehyde evaluated. For each trap type, bars not superscribed by the same letter are significantly different according to ANOVA ($P< 0.05$) and LSD tests.

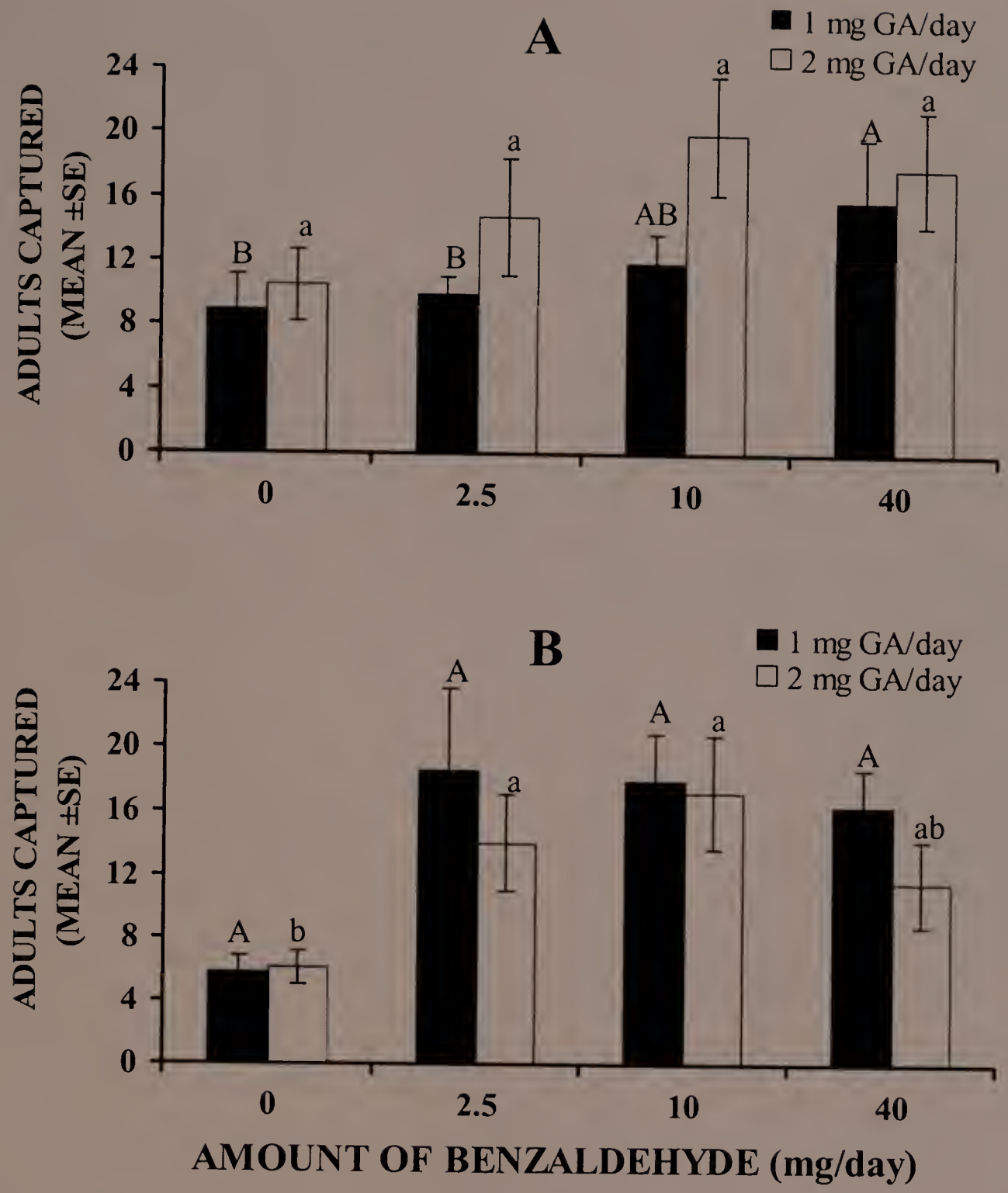

Figure 10. Plum curculio captures (mean ± SE) by A) panel traps and B) pyramid traps as a function of amounts of benzaldehyde released per day in 2002 in association with grandisoic acid (GA) at either 1 or 2 mg/day of release. For each trap type, bars not superscribed by the same letter are significantly different according to ANOVA ($P<$ 0.05) and LSD tests.

2003 Evaluation of Benzaldehyde

Results from this follow-up study (involving grandisoic acid released at 1 mg/day on average) showed that, for panel traps, plum curculio captures were significantly enhanced by the addition of benzaldehyde to grandisoic acid, irrespective of the amount of benzaldehyde released from vials (ANOVA df= 3, 9; F= 4.38; *P*= 0.04). For pyramid traps, the most attractive release rate of benzaldehyde was 10 mg/day; additional amounts of benzaldehyde did not increase, but rather significantly decreased, the attractiveness of benzaldehyde to plum curculios (ANOVA df= 3, 9; F= 6.95; *P*= 0.01) (Fig. 11).

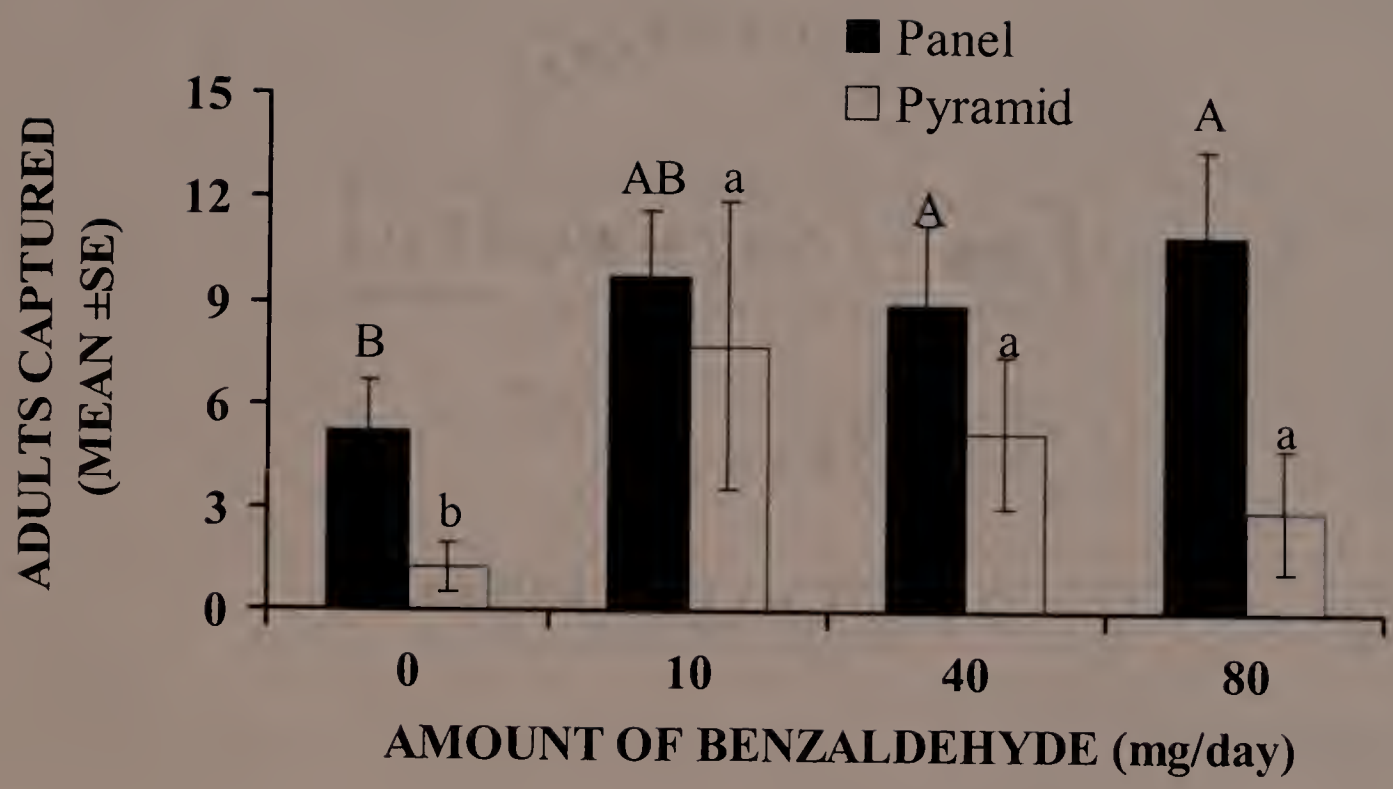

Figure 11. Plum curculio captures (mean ± SE) by panel and pyramid traps as a function of amounts of benzaldehyde released per day in 2003 in association with grandisoic acid at 1mg/day of release. For each trap type, bars not superscribed by the same letter are significantly different according to ANOVA ($P< 0.05$) and LSD tests.

Discussion

The 2001 results provided evidence that benzaldehyde, naturally present in both apple flowers (Buchbauer et al., 1993) and developing fruit (Leskey et al., 2001), synergistically enhanced the response of plum curculios to the male-produced pheromone grandisoic acid. This contrasted with results for ethyl isovalerate and limonene, which indicated that addition of either fruit volatile to grandisoic acid did not improve (at the dose evaluated) the attractiveness of grandisoic acid. In the 2002 study, benzaldehyde at 10 mg/day of release (particularly when in association with grandisoic acid at 1 mg/day of release) was the most attractive bait combination for plum curculios when considering both panel and pyramid traps. The 2003 study confirmed results of 2002 and additionally showed that benzaldehyde at 80 mg/day of release in association with grandisoic acid (1 mg/day of release) did not further enhance (for panel traps) and actually decreased (for pyramid traps) plum curculio captures.

This study is one of the few showing that a single synthetic component of host odor synergizes the attractiveness of an aggregation pheromone to a species of weevil without using supplementary sources (natural or synthetic) of host plant odor. Most other reports (e.g., Dowd and Bartelt, 1991; Bartelt et al., 1992; Lin et al., 1992; Weissling et al., 1994; Giblin-Davis et al., 1996; Hammack, 2001) show that a synthetic aggregation pheromone in combination with blends of synthetic and/or natural food odor is needed to synergistically increase attraction in beetles.

My 2001 findings indicate that (1) among all baits evaluated here, benzaldehyde+ grandisoic acid stands as the most attractive lure for plum curculios, whereas grandisoic

acid alone is mildly attractive and benzaldehyde alone is only weakly attractive, (2) the response of plum curculios to the combination of benzaldehyde + grandisoic acid was similar for panel and pyramid traps, which suggests that plum curculios are similarly attracted to this lure regardless of their mode of immigration (i.e., by flight or by crawling) into an apple orchard, and (3) benzaldehyde+grandisoic acid is highly attractive for immigrant plum curculios of both sexes (data not shown).

The type of interaction found between benzaldehyde and grandisoic acid in my 2001 study differs from that found in West Virginia for the bivoltine "southern strain" plum curculio (T.C. Leskey and S.E. Wright, unpublished data). In their study, involving the same kind of traps and odors, Leskey and Wright found that plum curculios responded equally to benzaldehyde alone and benzaldehyde in combination with grandisoic acid, and that grandisoic acid alone ranked third in attractiveness. Such contrasting results might be explained by the fact that adult plum curculios used in Leskey and Wright's study were dislodged from host trees (i.e., they had already immigrated into host trees) before testing, and therefore may have been in a different physiological state. Alternatively, there might be variation in responsiveness to these types of volatiles between the northern (e.g., Massachusetts) and southern (e.g., West Virginia) strains of plum curculio. Therefore, more detailed information about the role of benzaldehyde and GA in host-finding, feeding, mating, and oviposition for both plum curculio strains is needed.

Results from the 2002 experiment suggest that a low release rate of grandisoic acid (1 mg/day) is more cost-effective for use in both panel and pyramid traps than a higher release rate of grandisoic acid (2 mg/day).For panel traps, benzaldehyde

performed best at the high release rates (10 or 40 mg/day) regardless of the amount of grandisoic acid released from panel traps. For pyramid traps, however, the attractiveness of benzaldehyde decreased somewhat at the highest release rate, particularly when in association with grandisoic acid at the high release rate (2 mg/day), suggesting some degree of close-range repellency.

Results from my 2003 study indicated that for panel traps, benzaldehyde at 80 mg/day of release in association with grandisoic acid released at 1 mg/day did not enhance captures relative to lower amounts of benzaldehyde. Moreover, for pyramid traps benzaldehyde at high release rates influenced negatively the attractiveness of the binary combination of benzaldehyde+grandisoic acid, corroborating the close-range repellency found in our 2002 study.

My collective findings, together with an assessment of the cost of benzaldehyde (as formulated by us) and grandisoic acid (a commercial formulation), suggest that benzaldehyde at a release rate of 10 mg/day/trap, in association with grandisoic acid at a release rate of 1 mg/day/trap, seems to be the most cost-effective bait combination for use with both panel and pyramid traps to monitor adult plum curculios. An alternative use of this odor combination, at a release rate of benzaldehyde of 40 mg/day in association with grandisoic acid at 1 mg/day of release, has been put forward under a 'trap-tree' approach, which has proven useful for simplifying the monitoring of injury inflicted by plum curculio to fruit (Prokopy et al., 2003, R.J. Prokopy, unpublished data).

I conclude that use of panel and pyramid traps baited with the attractive lure benzaldehyde+grandisoic acid at proposed release rates could be helpful to apple growers for several reasons: (1) when deployed adjacent to forested areas of unsprayed orchards,

such traps have proven very useful in tracking the seasonal course of immigration of overwintered plum curculios for five consecutive years (chapter 4), (2) under this approach, traps intercept immigrant plum curculios before they reach host trees, and thus prior to their engagement in feeding, mating, and oviposition activities, which could change the degree of plum curculio responsiveness to lures, and (3) benzaldehyde+grandisoic acid remains highly attractive as a lure to plum curculios throughout the entire period of immigration (62 days in 2001, 82 days in 2002, 77 days in 2003) regardless of the stage of bud and fruit development and sex of adults.

Table 3. Type of interaction found between single attractants (synthetic fruit odor or GA) and combined attractants (synthetic fruit odor + GA) when evaluated in association with panel and pyramid traps for capturing plum curculios in 2001. BEN= benzaldehyde, GA= grandisoic acid (plum curculio pheromone), EIV= ethyl isovalerate, LIM= limonene.

Trap type	Chemical	Ratio of Interaction (ROI)[a]	Type of interaction
Panel	BEN+GA	1.58 ± 0.17*	Synergistic[b]
Pyramid	BEN+GA	1.65 ± 0.27*	Synergistic[b]
Panel	EIV+GA	0.88 ± 0.18	Additive
Pyramid	EIV+GA	0.90 ± 0.21	Additive
Panel	LIM+GA	0.66 ± 0.13*	Inhibitory
Pyramid	LIM+GA	0.90 ± 0.28	Additive

[a] Mean ± SE; *$P< 0.05$ for the null hypothesis that ratio of interaction (ROI) = 1 (Hammack, 1996), by a two-tailed t test ($df= 3$).

[b] As further verified by orthogonal contrast analysis ($P< 0.001$ and $P< 0.01$ for panel and pyramid traps, respectively).

CHAPTER 4

TEMPORAL DYNAMICS OF PLUM CURCULIO IMMIGRATION INTO AN APPLE ORCHARD IN MASSACHUSETTS

Abstract

I investigated, over a five-year period, temporal dynamics of plum curculio immigration into an unsprayed section of a commercial apple orchard in an attempt to establish the relationships between the timing and extent of immigration, weather factors and phenological tree stage. By using panel and pyramid traps baited with synthetic attractive odor and deployed near woods adjacent to orchard trees, I exploited some of the chemical cues potentially initiating and directing the spring immigration by plum curculios. Traps were inspected on a ~daily basis over the entire period of immigration. Across all five years, 59% of the potential immigrants arrived at host trees by petal fall, as determined by trap captures. Based on my combined data, I propose the occurrence of pre- and post-petal-fall periods of plum curculio immigration, each of which is influenced to a different extent by temperatures prevailing in spring. For each of the five trapping years, the influence of daily air temperature on captures by either panel or pyramid traps was greater during the pre-petal-fall period than the post-petal-fall period. Thermal constants (expressed in Degree Days [DD] base 6.1°C starting January 1st) for the start of plum curculio immigration, and for the 50th and 80th percentiles of cumulative captures

were 113, 249 and 412 DD, respectively. Pesticide application procedures are recommended to maximize plum curculio control while minimizing exposure and cost.

Introduction

A fundamental step in the formulation of pest management strategies is the development of effective monitoring tools, a process that ultimately depends on a comprehensive understanding of the ecology and behavior of the insect pest (Foster and Harris 1997). For instance, an important aspect of pest behavior that can be exploited by pest management practitioners is the manner by which a pest individual approaches natural resources, namely food, mates or egg-laying sites (Haussman et al. 2004). The type and timing of movement into an area (immigration) and out of an area (emigration) by a pest individual is also vital to understanding the dynamics of the pest population (Pedigo 2001).

The plum curculio, *Conotrachelus nenuphar* (Herbst) (Coleoptera: Curculionidae), has been regarded customarily by most commercial fruit growers in eastern and central North America as one of the most injurious insect pest of apples (Chouinard et al. 1992a, Vincent et al. 1999). Damage to fruit results from feeding and oviposition scars produced by adult females and from burrows by larvae. Oviposition damage is economically more important than feeding damage (Racette et al. 1992).

Many aspects of plum curculio ecology and behavior are well known, as they have been worked out carefully by many researchers. In northeastern North America, adults overwinter in a state of reproductive diapause, which terminates the following

spring (Smith and Flessel 1968). Forested areas adjacent to orchards are the primary overwintering sites, but some overwintering can take place within orchard blocks (Lafleur et al. 1987, Chapter 5). McGiffen and Meyer (1986) previously determined that post-diapause development of adult plum curculios is completed before the end of winter, and that adult activity and dispersal in the spring are largely suppressed by low temperatures.

The timing of emergence of plum curculios from overwintering habitats and their immigration into apple orchards has most often been related to either air and soil temperatures or to host plant phenology. However, the relative influence of abiotic and biotic factors on the timing and extent of plum curculio immigration has not been quantified before. Temperature has been treated as the primary abiotic factor triggering plum curculio emergence and immigration into orchard trees by most researchers (e.g., Quaintance and Jenne 1912, Chapman 1938, Bobb 1949, Smith and Flessel 1968, McGiffen and Meyer 1986). In contrast, others (e.g., Whitcomb 1929, Armstrong 1958) have related the seasonal abundance of plum curculios to particular stages of tree phenology.

Determining need for and timing of insecticide applications that will protect fruit from injury by plum curculio that are based on presence of adults on host trees has been a critical aspect for managing populations adequately (Racette et al. 1992, Vincent et al. 1999, Prokopy et al. 2003, 2004). In concept, a reduction in amount of insecticide used against plum curculio from the current norm of three spray applications during May and June (Reissig et al. 1998) to an amount that is precise according to need should be accompanied with an effective approach to monitoring the course of plum curculio

immigration. Limb jarring, an approach that involves tapping tree limbs using a pole to dislodge plum curculios onto an underlying ground cloth, is one of the methods traditionally used to determine the time of first appearance, location, and relative abundance of plum curculios within an orchard (Quaintance and Jenne 1912, Chapman 1938, Racette et al. 1992). However, limb jarring has several shortcomings: (1) it is labor intensive; (2) it can dislodge blossoms or fruit; (3) its efficacy is highly dependent upon tree size, weather, time of day, and other factors; (4) it cannot be used to study immigration because plum curculios that are able to overwinter within orchards will be confounded with true immigrants; and (5) the difficulty associated with sampling at night, the time when plum curculios are most active on trees (Racette et al. 1991; Chouinard et al. 1992b, 1993, 1994).

An alternative for monitoring the onset and extent of plum curculio immigration into apple orchards is the utilization of traps baited with synthetic attractive odor and deployed in close proximity to the forested areas that are the main overwintering sites. Here, I investigated, over a five-year period, the temporal dynamics of plum curculio immigration into an unsprayed section of a commercial apple orchard in Massachusetts using two different trap types baited with attractive synthetic odors. In particular, I aimed at (1) characterizing the overall pattern of immigration from woods into an apple orchard by overwintered plum curculios; (2) determining the relationships among trap captures, tree phenology and weather; (3) estimating thermal constants for different stages (onset, 50^{th} and 80^{th} percentiles of cumulative captures) of plum curculio immigration; and (4) determining the relative predictability of different stages of plum curculio immigration by comparing tree phenology versus thermal constants.

Materials and Methods

This study was conducted over a period of five years (2000-2004) at the University of Massachusetts Cold Spring Orchard (Belchertown, Massachusetts). I used a ~1 ha unsprayed block comprised of a larger section having 216 small (M.9 rootstock) trees (primarily McIntosh and Red Delicious) located on the eastern side, and two smaller sections, located on the western side, having 145 medium-sized (M.26 rootstock) trees of various disease-resistant cultivars (Fig. 12). The inter-row and inter-tree distances were 5 m and 4 m, respectively, for trees on M.9 rootstock, and 6 m and 4.5 m, respectively, for trees on M.26 rootstock. The perimeter of the entire block, bordered almost entirely by mixed deciduous forest, was about 500 m.

Traps used in the study were of two different types: (a) clear sticky Plexiglas panels (60 by 60 cm), intended to capture adult plum curculios in flight, and (b) trunk-mimicking black pyramid traps, (designed originally by Tedders and Wood [1994] to monitor pecan weevils), which capture plum curculios approaching host trees primarily by crawling. The woods-facing side of each panel was coated with Tangletrap glue (Orchard Equipment Supply Company Inc., Conway, MA) to capture plum curculios that were presumably immigrating from the woods into the orchard block. Trap design is described in more detail in Chapter 2.

For each of the five years, traps were deployed in pairs (one trap of each type spaced 1 m apart) along the periphery of the orchard, in close proximity (~1 m) to the woods, under the assumption that the majority of plum curculios captured by traps had overwintered in the forest and were immigrating into the orchard block. Each pair of

traps was spaced 10 m from other trap pairs on either side except in 2004, when the distance between each trap pair was 35 m. For each of the five years, trap captures were pooled across all traps of the same type deployed in the orchard. The predominant bait used for luring plum curculios to traps was composed of benzaldehyde, a synthetic component of host plant odor, and grandisoic acid, the plum curculio aggregation pheromone. When in combination, these two odors constitute a very attractive lure for adult plum curculios (Chapters 2 and 3).

Trap deployment and baiting took place during the silver-tip stage of bud development on each trapping year. Traps were inspected for plum curculio captures on a ~daily basis (7:30-10:00 AM) from trap deployment until fruit reached 3 cm in diameter (by late June/early July). All adult plum curculios captured were sexed in the laboratory.

Data Analysis

The process of immigration into the apple orchard was characterized by the day of first captures in traps and days of the 50^{th} and 80^{th} percentiles of cumulative captures. The latter occurred around petal fall, the stage of tree phenology at which plum curculios exhibit the highest activity and dispersal (Lafleur and Hill 1987), and the time at which the first insecticide is applied against plum curculio in Massachusetts orchards (Prokopy et al. 2003, 2004). I ended the studies by late June/early July, when no captures occurred for 3-4 consecutive days with relatively high temperatures. A Kolmogorov-Smirnov test was used to determine non-parametrically, using pairwise comparisons, whether the cumulative distributions of captures varied across years. Because Kolmogorov-Smirnov

tests are sensitive to differences in the general shapes of the distributions in two samples only, then I decreased the *P*-value accordingly using Bonferroni correction to account for the ten different pairwise comparisons being performed (Bonferroni, 1936, Rice 1989). Whether the median of plum curculio captures was the same across the five trapping years was determined also non-parametrically via Kruskal-Wallis tests.

The numbers of plum curculios captured by panel and pyramid traps before and after petal fall were compared across years using a Chi-square to test the null hypothesis that the two criteria of classification (captures before vs. after petal fall, and year) were independent from each other. Tree phenology was monitored on a daily basis on the MacIntosh trees, the cultivar most commonly used for monitoring apple tree phenology in New England (ProNew England Apple Crop Profile 2003). Differences in stage of tree phenology among the cultivars present in the orchard block were considered to be minimal. The following numerical code, modified from Chapman and Catlin (1976), was used to record the different stages of tree phenology: (1) silver tip, (2) green tip, (3) half-inch tip, (4) tight cluster, (5) first pink, (6) full pink, (7) first bloom, (8) full bloom, (9) petal fall, (10) within a week after petal fall, and (11) 2-6 wks after petal fall (depending on the year). Stages 1-9 were considered as pre-petal fall, whereas stages 10-11 were post-petal fall.

Weather data were taken at a weather station located 150 m from the orchard block and averaged on an hourly basis by data loggers (model CR10-X; Campbell Scientific Inc., North Logan, UT). To determine type and strength of relationships existing between daily trap captures and mean air temperature for plum curculio captures occurring either before petal fall (i.e., from the moment of first captures until the last day

of petal fall) or after petal fall, Pearson's correlation analyses (Pearson 1896) were conducted on each year's data separately for each trap type. Influence of weather factors, including temperature, on plum curculio captures for the year 2000 has been reported in Chapter 2, although under a different approach (i.e., in Chapter 2, the influence of weather factors on plum curculio captures was determined for 39 days without taking into account plum curculio captures before and after petal fall).

Thermal constants for the initiation of plum curculio immigration (START), 50th and 80th percentiles of cumulative captures were estimated using a temperature threshold of 6.1°C for resumption of adult activity after post-diapause development (McGiffen and Meyer, 1986). This threshold has also been used with the apple blossom weevil, *Anthonomus pomorum* (L.), (Ctvrtecka and Zdàrek 1992), and the boll weevil, *Anthonomus grandis* Boheman (Parajulee et al. 1996). On each trapping year, Degree Days (DD) started to accumulate on January 1st.

To determine whether there was more variability around the mean values of the thermal constants estimated and of tree phenology for the onset of plum curculio immigration across the five trapping years, I constructed two Coefficients of Variation (C.V.). The first C.V. involved mean thermal constants (using the average and Standard Deviation [S.D.] values obtained across five years), whereas the second C.V. involved the particular phenological tree stage at which plum curculio started immigrating into the orchard block (using the average and S.D. of the numerical code used on each year). The degree of correlation between tree phenology and thermal constants across the five trapping years was estimated using Spearman rank *R* correlation analyses for (1) the day

of first captures and (2) the 50th percentile of cumulative captures. All statistical analyses were performed using the software Statistica® (StatSoft 2001).

Results

Overall Pattern of Plum Curculio Immigration

In all, 4,279 plum curculios were captured by traps across all five trapping years (Table 4). On average, the period of plum curculio immigration lasted 63 days (± 6.0 SE), with the shortest and longest periods encompassing 51 days in 2000 and 85 days in 2002, respectively. Maximum captures occurring on a single day averaged 12.3% (± 5.8 SE) of the seasonal total, with values ranging from 6.9% (2004) to 21.6% (2000). As revealed in Fig. 13, the earliest start of immigration occurred in 2002 (on 14 April), whereas the latest start of immigration took place in 2001 (on 2 May). The relative distributions of captures (Fig. 13) and the median Julian day of captures varied significantly across years as determined by Kolmogorov-Smirnof tests (in all pairwise comparisons $P< 0.01$, after Bonferroni corrections) and Kruskal-Wallis (Chi-square= 186.3; df= 4; $P< 0.001$).

Plum Curculio Captures According to Tree Phenology

Plum curculios started immigrating when trees were either at the silver-tip stage (stage 1) (in 2004), at the tight-cluster stage (stage 4) (in 2000, 2002 and 2003) or at the first-pink tree stage (stage 5) (in 2001). Fifty-percent cumulative captures occurred when

trees were either in full bloom (stage 8) in 2000 and 2001, by petal fall (stage 9) in 2003 and 2004, or during the first week of fruit development (stage 10) in 2002. Eighty-percent cumulative captures took place during stage 10 (i.e., first week of fruit development) in four of the five years (2000-2003) or during stage 11 (i.e. after one week of fruit development) in 2004.

Table 4 shows that of the total number of plum curculio immigrants captured by panel and pyramid traps combined (potentially colonizing host trees), on average 59% had already been trapped by petal fall, with the remaining 41% being captured by traps after petal fall. A Chi-square test revealed that numbers of adults being captured by panel and pyramid traps before and after petal fall differed significantly across years (Chi-square= 122.9; df= 9; $P< 0.001$ for panel traps, and Chi-square= 72.7; df= 9; $P< 0.001$ for pyramid traps). The period of time required from the last day of petal fall to achieve 80% cumulative captures was one week in 2000 and 2004, two weeks in 2003, and three weeks in 2001 and 2002.

Relationship among Trap Captures, Tree Phenology and Weather

In general, captures by pyramid traps were less influenced by temperature than were by panel traps. There was a strong positive influence of mean daily air temperature on plum curculio captures by panel traps before petal fall for each of the five trapping years. In contrast, after petal fall the relationships between mean air temperature and captures by either panel or pyramid traps were rather weak except in 2003 for panel traps. In 2003, after-petal-fall captures by panel traps were positively correlated with

temperature (Fig. 14B). The proclivity of adults to either fly or crawl was independent of sex.

Thermal Constants for Different Stages of Plum Curculio Immigration

Table 5 shows the thermal constants (base 6.1°) for the different stages of plum curculio immigration. On average, immigration started when 112.6 DD (± 7.7 SE) had accumulated since January 1st. For the 50th and 80th percentiles of cumulative captures, the number of DD accumulated since January 1st were 248.7 (± 15.1 SE) and 412.5 (± 13.7 SE), respectively.

Relative Predictability of Plum Curculio Immigration: Tree Phenology versus Thermal Constants

I found less variability among years for the initiation of plum curculio immigration as determined by accumulation of DD (C.V. = 15.2) than by tree phenology (C.V. = 42.2). Spearman correlation analyses show that, across all five trapping years, values assigned to characterize tree phenology were not associated with thermal constants for the days of first captures (Spearman R= -0.22; P= 0.72). In contrast, thermal constant values estimated for the 50th percentile of cumulative captures were significantly and positively associated with phenological tree stage (Spearman R= 0.95; P= 0.01) (Fig. 15).

Figure 12. Unsprayed section of the apple orchard (UMASS Cold Spring Orchard; Belchertown, MA). Depending on the year, 14-48 panel (⊤) and 14-48 pyramid (▲) traps were deployed in pairs along the periphery of the orchard block, in close proximity to woods, the main overwintering sites of adult plum curculios. The distance among trap pairs was either 10 m (2000-2003) or 35 m (2004). The perimeter of the block was about 500 m.

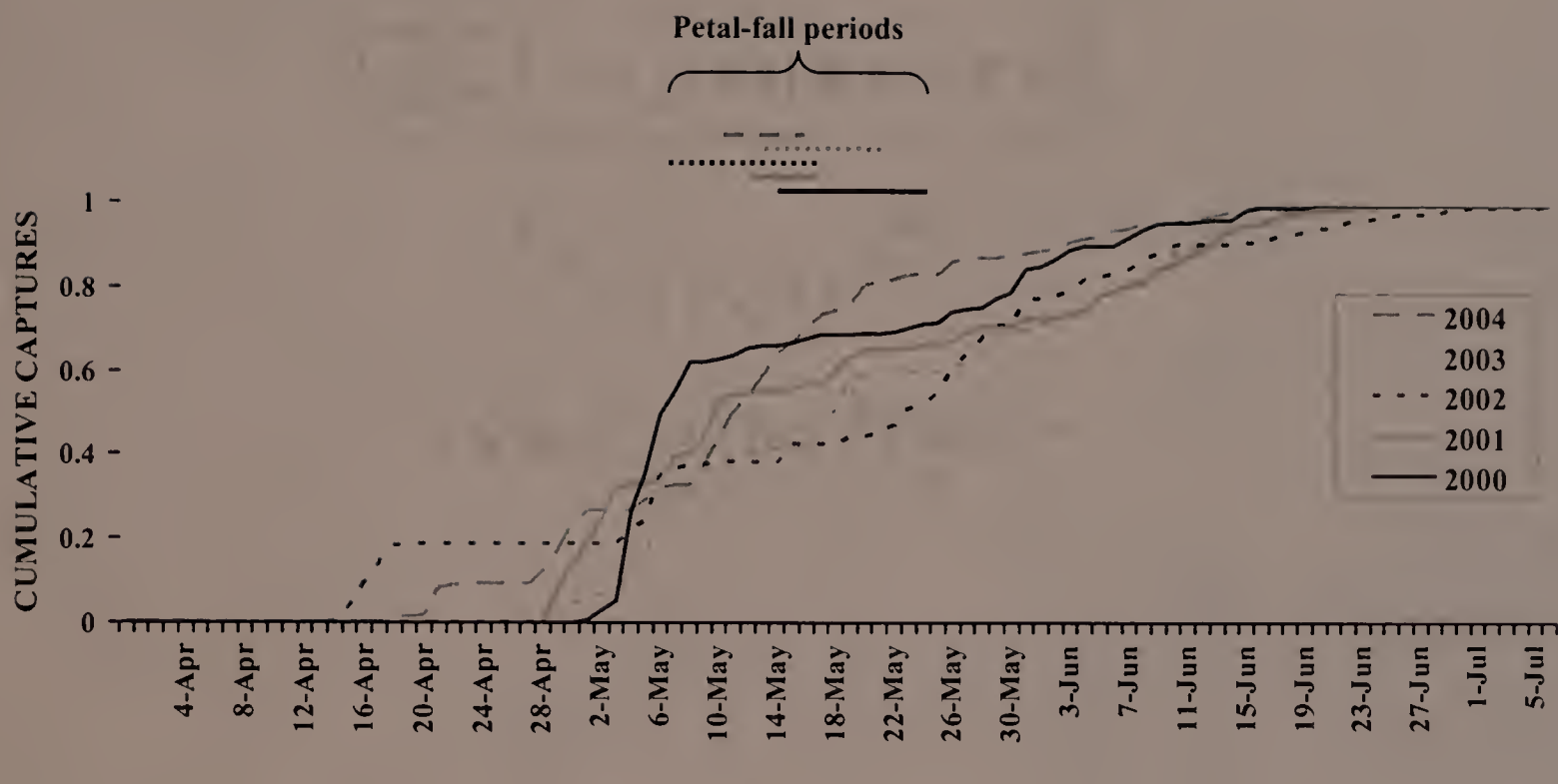

Figure 13. For each of the five trapping years, cumulative plum curculio captures (expressed as proportion of the total) by panel and pyramid traps combined, according to date. Lines above the graph indicate, for each year, the duration of the petal-fall period (the color and dash style of lines correspond to those used in the graph labels).

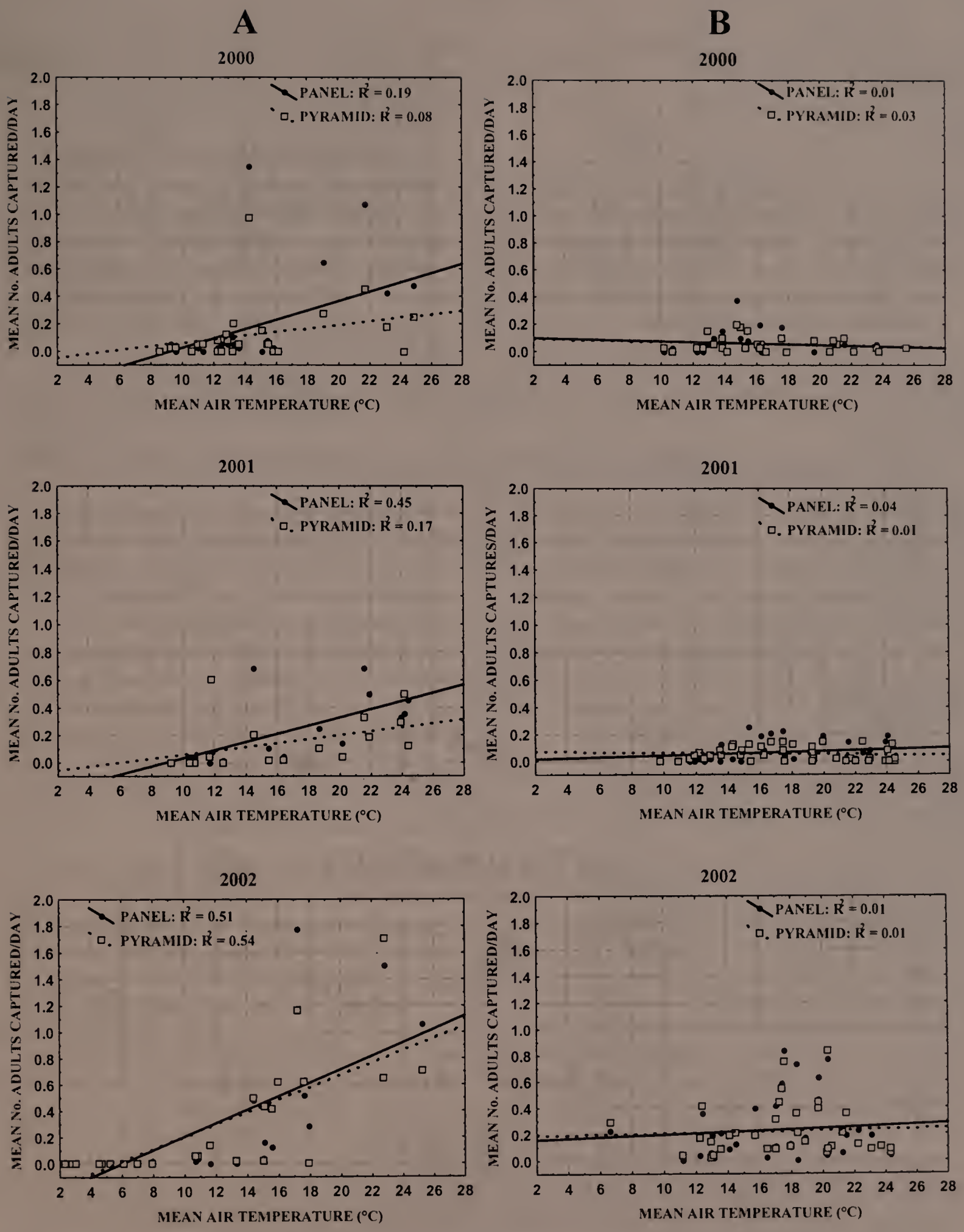

Figure 14. For each of the five trapping years, relationships between daily captures by panel and pyramid traps and mean daily air temperature, either A) before or B) after petal fall. The number of days before/after petal fall included in the correlation analyses was 23/52 in 2000, 17/43 in 2001, 33/51 in 2002, 24/30 in 2003, and 29/37 in 2004, respectively. Continued, next page.

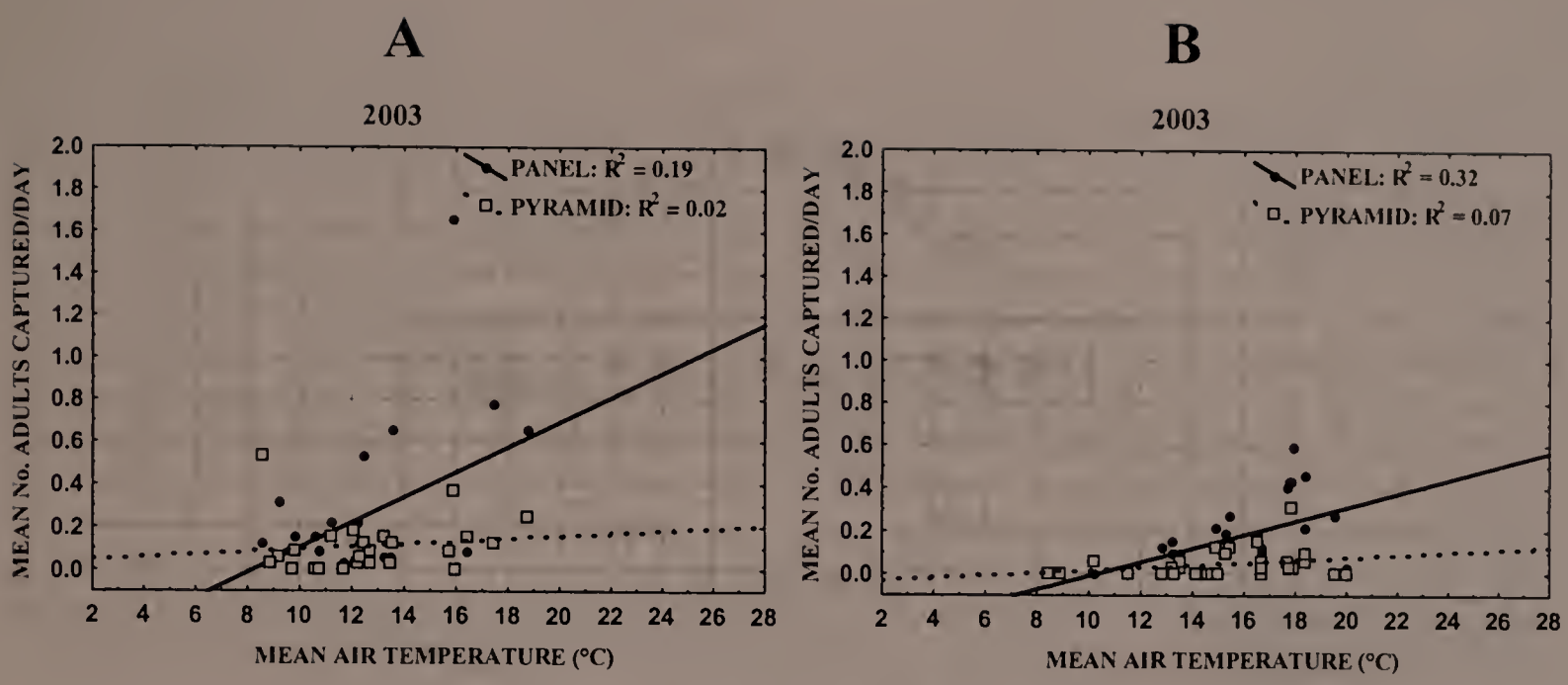

Figure 14. CONT. For each of the five trapping years, relationships between daily captures by panel and pyramid traps and mean daily air temperature, either A) before or B) after petal fall. The number of days before/after petal fall included in the correlation analyses was 23/52 in 2000, 17/43 in 2001, 33/51 in 2002, 24/30 in 2003, and 29/37 in 2004, respectively.

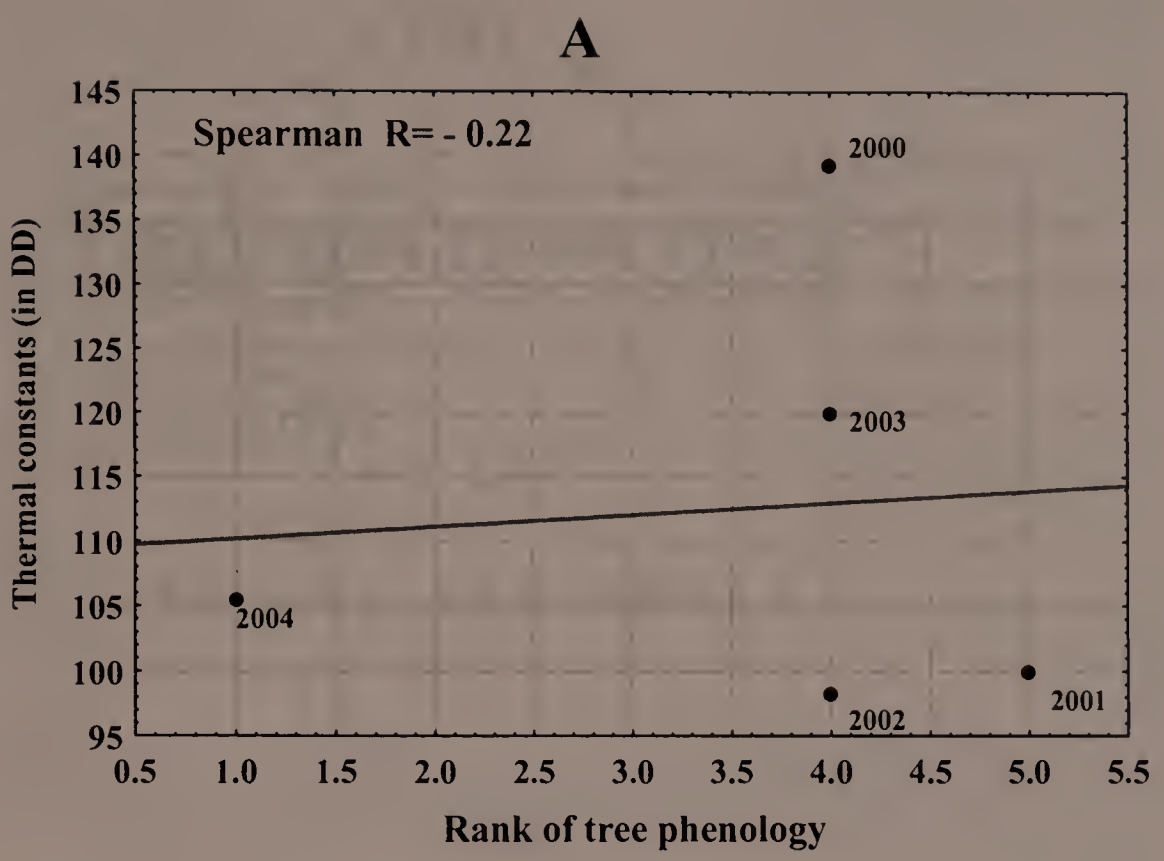

Figure 15. Relationships between thermal constants (expressed in Degree Days [DD], accumulated starting on Jan 1st) and stage of tree phenology (using a numerical code), for either (A) start of plum curculio immigration, or (B) 50th percentile of cumulative captures.

Discussion

This five-year study clearly shows that air temperature plays a key role in the process of orchard colonization by overwintered plum curculios in spring. Two of the most relevant findings are: (1) that the influence of temperature on patterns of plum curculio immigration is very strong during the pre-petal-fall period of immigration, whereas immigration taking place during the post-petal-fall period depended to a lesser extent on prevailing temperatures, and (2) that the relative predictability of the onset of plum curculio immigration could be explained better in terms of accumulation of Degree Days (base 6.1°C and computed starting January 1st) rather than by tree phenology.

In this study, I focused on the process of adult immigration from overwintering sites (which primarily are woods) rather than adult activity on host trees. This is important to mention because, upon reaching host trees, the duration of time spent on host trees and the different types of behavior (e.g., crawling, mating) exhibited by adult plum curculios have been found to depend primarily on host plant phenology (Smith and Flessel 1968, Owens et al. 1982, Racette et al. 1991, 1992, Chouinard et al. 1993, 1994), probably because of their imperative need to find potential mates as well as feeding and oviposition sites. Consequently, because odor-baited traps were deployed along the periphery of the orchard block and inspected on a ~daily basis for the entire period of immigration, I believe this study examined timing and extent of plum curculio immigration from woods more accurately than previous studies that have relied on branch-tapping (which yields adults present already on host trees). As stated in the Introduction of this chapter, branch-tapping, the most widely used method for seasonal

monitoring of adult activity and abundance, beside its inaccuracy, is also likely to yield plum curculios that may have overwintered inside orchard blocks (Chapter 5).

In temperate regions, where temperatures rise relatively rapidly and predictably in spring, a simple threshold temperature can reliably time the onset of activity to coincide with favorable conditions (Danks 1987). My empirical Degree-Day model predicts the start of adult plum curculio immigration at about 113 DD (base 6.1°C). Previously, Lafleur and Hill (1987) determined in Quebec that 330 DD (base 0°C) had accumulated by the tight-cluster tree stage, the stage at which the first peak of immigration was observed. In this study I also determined (results not shown) that less heat was accumulated for the start of immigration (~290 DD base 0°C) in Massachusetts than in Quebec when I used 0°C as a post-diapause developmental threshold. This difference in amount of heat required by overwintered plum curculios in Massachusetts vs. Quebec may be probably due to, at least in part, differences in the severity of the winter months in both regions, as adult plum curculios from both regions show similar supercooling points (Ch. Vincent, unpublished). Degree-day-based models are also being evaluated for the southern race of plum curculio to correctly time scouting and/or trapping (Mulder et al. 1997, Lan et al. 2004).

Temperature seems to be the primary environmental cue for the termination of diapause itself in many insect species inhabiting temperate regions (Danks 1987). The timing of immigration of overwintered plum curculios had been related to either soil and air temperature or to host plant phenology, but the relative influence of these two environmental factors had not been quantified in detail. Here, I found that, for the onset of plum curculio immigration, there was less variability among the five trapping years for

the estimation of DD (base 6.1°C and accumulated starting on Jan 1st) than for the stages of tree phenology, which showed more variability as determined by the CVs estimated. This result was confirmed by the lack of correlation existing between DD and stage of tree phenology. Thus, examination of stages of bud development in spring as a monitoring tool for forecasting timing of plum curculio immigration can be considered as unsuitable. Such a lack of synchronization between start of spring dispersal by plum curculios and tree phenology may be due to a complex effect of soil temperature on apple bud development, which might be different from the actual temperatures experienced by adult plum curculios in their overwintering microhabitats (Toepfer et al. 2002). In the plum curculio, diapause is completed during the winter months, before favorable conditions return in spring (McGiffen and Meyer, 1986). Therefore, adult plum curculios seem to respond more rapidly than their host plants to changes in temperature. Analogous results have been reported with the apple blossom weevil, *A. pomorum*, in Switzerland (Toepfer et al. 2002), a species that shares several biological and ecological attributes with the plum curculio.

Evidently, there are several environmental factors potentially influencing plum curculio immigration other than temperatures prevailing in spring. These include visual (e.g., tree trunk silhouette [Butkewich and Prokopy 1997, Leskey and Prokopy 2002]) and chemical cues (e.g., host-derived volatiles, aggregation pheromones emitted by males [Chapters 2 and 3, Eller and Bartelt 1996, Leskey and Prokopy 2000, Leskey et al. 2001]) that aid plum curculios to locate host trees and thereby potential mates, food and oviposition sites.

From the practical point of view, the trap-capture patterns obtained over a five-year period allow us to characterize plum curculio immigration as follows: First, stretches of hot weather occurring during the pre-petal-fall period (as in our 2000 season) are conducive to concentrated emergence and immigration of plum curculios. Under these conditions, most adults may be present within orchards before the end of the pre-petal-fall period. Thus, in years with high pre-petal-fall temperatures a petal-fall spray covering the entire orchard block is recommended and should yield excellent control of the bulk of the population. Second, during the post-petal-fall stage plum curculio immigration into the apple orchard continues but with a lesser influence of weather, unless cool temperatures (such as in our 2002 season) have prevailed during the pre-petal-fall period, which would lead to an extended period of plum curculio emergence and immigration. Using similar reasoning, Reissig et al. (1998) developed an oviposition model involving accumulation of 171 DD (base 10°C) after petal fall to predict for how long insecticide should be applied to orchard trees to prevent injury by plum curculio late in the season. Third, during and about two weeks after petal fall a proportion (yet not determined) of plum curculios may actually be re-colonizers rather than immigrants as indicated, among other evidence, by substantial back-and-forth movement by adult plum curculios between woods and orchard trees (Piñero and Prokopy 2005). Under this scenario, damage to fruit sampled at harvest by plum curculio in Quebec and other areas of the northeast occurring after fruit set (Lafleur and Hill 1987, Chouinard et al. (1992a) may be a consequence of re-infestations that occurred after the petal-fall spray of insecticide.

I recommend that, depending on the type of weather (primarily temperature) prevalent during the pre-petal-fall period of plum curculio immigration, the first spray of

insecticide (commonly applied by growers by the time of petal fall) should be delayed either (1) for one week if the pre-petal fall period of immigration is characterized by high temperatures (as in our 2002 season), or (2) by 10-14 days if cool, rainy weather prevails during the pre-petal fall stage of immigration. By doing this, a grower could maximize plum curculio control as a higher proportion of immigrants may be killed, while costs and exposure to insecticide would be minimized, as fewer applications of insecticide might be needed across the entire season. This is analogous to the temperature model developed at Cornell University by Reissig et al. (1998) to control plum curculio, based on the type of weather prevailing after petal fall.

I conclude that (1) odor-baited traps can be used to accurately track plum curculio immigration into sprayed commercial orchards, particularly in the absence of another monitoring tactic for plum curculio, such as an odor-baited tree approach (Prokopy et al. 2003, 2004), and (2) prediction of the timing of plum curculio immigration can be achieved using cumulative Degree Days (base 6.1°C and computed January 1st); however, we recommend that thermal constants reported here be validated in additional locations in the Northeast.

Table 4. For each of the five trapping years, plum curculio captures by panel and pyramid traps before petal fall (i.e., phenological tree stages 1-9) and after petal fall (i.e., phenological tree stages 10-11).

EVENT	2000	2001	2002	2003	2004	Avg ± SE
Total no. adults captured	430	544	1,354	485	1,366	
Last day of petal fall	05/24	05/16	05/17	05/22	05/14	
Cum. captures by panel traps[1]	185	182	290	202	312	
Cum. captures by pyramid traps[1]	122	121	285	87	565	
Cum. captures (panel+pyramid)[1]	307	303	575	289	877	
% of total[1]	**71.4**	**55.7**	**42.5**	**59.6**	**64.2**	**58.7 ± 4.8**

[1]captures up to last day of petal fall

Table 5. For each of the five trapping years, date and stage of tree phenology for the first captures, and thermal constants (expressed in Degree Days [DD]) estimated for different stages of plum curculio immigration (START, 50th and 80th percentiles of cumulative captures). See Materials and Methods for a description of the numerical codes used to characterize phenological tree stage.

EVENT	2000	2001	2002	2003	2004	Mean ± SE
START (date)	05/02	04/30	04/15	04/29	04/17	
START (rank of tree phenology)	(4)	(5)	(4)	(4)	(1)	3.6 ± 0.7
START ($DD_{6.1°C}$)	139.3	99.9	98.3	119.9	105.5	112.6 ± 7.7
50th percentile (date)	05/07	05/11	05/24	05/19	05/12	
50th percentile ($DD_{6.1°C}$)	206.9	232.2	291.2	238.8	274.3	248.7 ± 15.1
80th percentile (date)	06/01	06/08	06/05	06/08	05/21	
80th percentile ($DD_{6.1°C}$)	418.3	456.4	420.7	388.9	378.5	412.5 ± 13.7

CHAPTER 5

EARLY-SEASON DISTRIBUTION AND EXTENT OF OVERWINTERING BY ADULT PLUM CURCULIOS WITHIN COMMERCIAL APPLE ORCHARDS IN MASSACHUSETTS

Abstract

I determined the early-season distribution of adult plum curculios, *Conotrachelus nenuphar* (Coleoptera: Curculionidae), within apple orchard blocks in Massachusetts in relation to tree size, as well as the extent to which adults were able to overwinter with respect to the type of weed management. Studies were conducted over a period of two years using Circle and pyramidal emergence traps under both sprayed and unsprayed conditions. In sprayed orchard blocks having large trees (M.7 rootstock), I determined that, by petal fall, most plum curculios were found on perimeter-row trees, as determined by Circle trap captures. In contrast, more plum curculios were found inside of blocks rather than on perimeter-row trees in blocks having small, full dwarf trees (M.9 rootstock). Adult plum curculios were capable of overwintering inside orchard blocks in substantial numbers. More plum curculios were found overwintering beneath perimeter-row trees in the plot not subjected to weed management than in the managed plot. My combined findings support the current recommendation of a whole-block insecticide

spray against plum curculio by the time of petal fall in Massachusetts orchards, and may call for a second whole-block application if blocks have small trees.

Introduction

The plum curculio, *Conotrachelus nenuphar* (Herbst) (Coleoptera: Curculionidae), is an important pest of pome and stone fruit in eastern and central North America (Racette et al. 1992, Vincent et al. 1999). One of the greatest challenges traditionally faced by apple growers in the northeast is the determination of need and timing of insecticide applications aimed at protecting fruit from injury by plum curculio. Consequently, since the first report published by Riley (1871), numerous studies have been conducted to determine not only the timing of spring emergence of adult plum curculios but also within-orchard adult dispersal and patterns of plum curculio behavior, in an attempt to use this information for the development of reliable monitoring and/or control methods (reviewed by Quaintance and Jenne 1912, Holloway 1977, Racette et al. 1992, Vincent et al. 1999).

Given the cryptic appearance, crepuscular habits, and thanatosis (i.e., dropping) behavior of adult plum curculios (Quaintance and Jenne 1912, Chapman 1938, Owens et al. 1982, Chouinard et al. 1993), studies on the spatial and temporal patterns of adult plum curculio overwintering and immigration into apple orchards have been conducted mainly using radioactively-labeled adults (e.g., Lafleur and Hill 1987, Lafleur et al. 1987, Chouinard et al. 1992) or by tapping fruit limbs with a pole to dislodge adult plum curculios and capturing them on a ground cloth (Quaintance and Jenne 1912, Chapman

1938). At the orchard level, combined findings have shown that (1) in the fall, most adults leave the orchards seeking overwintering sites in the woods, although a small proportion of plum curculios may overwinter within Quebec orchards depending on the microhabitat (Lafleur et al. 1987); (2) about two weeks after petal fall, most immigrant plum curculios are already present on orchard trees (Whitcomb 1929, Chapman 1938, Lathrop 1949, Blanchet 1987, chapter 4); and (3) upon reaching orchard trees, plum curculios show a tendency to stay on perimeter-row trees (Quaintance and Jenne 1912, Rings 1952, Chouinard et al. 1992); however, as the season progresses, some individuals may move from perimeter-row trees toward interior trees (Lafleur and Hill 1987, Chouinard et al. 1992). Because damage to fruit by plum curculio is commonly greater on peripheral trees of an apple orchard block than on interior trees (Chapman 1938, Chouinard et al. 1992, Vincent et al. 1997), current approaches to controlling plum curculio effectively in Massachusetts orchards involve application of a whole-orchard insecticide spray at or soon after petal fall, followed by one to three insecticide applications confined exclusively to peripheral-row apple trees (Chouinard et al. 1992, Prokopy et al. 2003a).

Adult plum curculios present on perimeter-row apple trees early in the spring may be either immigrants that moved from forested areas or individuals that overwintered inside orchard blocks. Whether tree size is a factor influencing the propensity of plum curculios to be present mainly on perimeter-row trees early in the season is an aspect that has not been evaluated before under field conditions. Similarly, only Lafleur et al. (1987) have quantified, in Quebec apple orchards, the extent of overwintering by adult plum curculios beneath perimeter-row trees in relation to the type of weed management. Here,

I aimed at determining, under the conditions of Massachusetts apple orchards, (1) the influence of tree size on the extent to which overwintered adult plum curculios penetrate into interior rows of sprayed sections of commercial orchard blocks before petal fall, and (2) the extent to which plum curculios are able to overwinter inside sprayed and unsprayed sections of commercial orchards (more specifically beneath perimeter-row trees), and whether the type of weed control influences the outcome.

Materials and Methods

Orchard Penetration Study

This study was conducted during April and May of 2003 and 2004 in eight commercial apple orchards in Massachusetts. Within each orchard, selected blocks had similar length (about 180 m of perimeter-row trees) and depth (at least 60 m). Five orchards in 2003 and four orchards in 2004 had large (M.7 rootstock) trees; the remaining orchards had small (M.9 rootstock) trees. All trees within a block were of the same size. Blocks having small trees were either of the same cultivar (primarily Gala), except one block that had alternate rows of Gala and Empire. Likewise, blocks having large trees were of the same cultivar (MacIntosh), except one block that had alternate rows of Red Delicious and Cortland. The average heights of large and small trees were 3.9 m (± 0.2 SE; n= 8) and 2.7 m (± 0.1 SE; n= 17), respectively. Accordingly, the average tree canopy radii were 2.4 m (± 0.1 SE; n= 16) for large trees and 0.9 m (± 0.04 SE; n= 25) for small trees.

Circle traps, made of aluminum screen with a plum-curculio-capturing device integrated on top, were used for the orchard-penetration study. This trap, originally developed for pecan weevil in Oklahoma (Mulder et al. 1997), was wrapped around the base of trees so as to completely encircle the trunk, thereby intercepting adults walking upward. For each block, 20 Circle traps were deployed at the green-tip tree stage (on April 24 and April 18, for 2003 and 2004, respectively) on trees located in the central part (about 60 m from the perimeter) of each orchard block. Use of the central section only (comprising 60 m) within a block minimized potential penetration of plum curculios from the lateral or back sides. For each block, traps were arranged in four transects of five traps each (denoted as A-E in Fig. 16), starting on perimeter-row trees. Because there were different inter-row distances due to the two different tree sizes, blocks having large trees received traps deployed in consecutive rows (1-5) (Fig. 16) and blocks having small trees received traps deployed in rows 1, 4, 7, 10, and 13 (Fig. 16B) for each of the four transects per block. Under this layout, traps were deployed at similar distances inside a block: on perimeter-row trees (a), and on trees located about 10-12, 20-22, 30-32 and 40-42 m inside of perimeter-row trees (denoted respectively as b, c, d, and e in Fig. 16A, B).

In 2003, for two of the four transects in each block, traps were baited with one dispenser of plum-curculio pheromone (releasing 1 mg/day of grandisoic acid) in association with one dispenser releasing the attractive host plant odor benzaldehyde (Chapter 3) at a low release rate (2.5 mg/day). The rationale for baiting these traps was to maximize the likelihood of capturing some plum curculios, as population densities were low in the sprayed orchard blocks in 2002. Traps for the two remaining transects per block were left unbaited. Because baiting half of the traps in 2003 was not associated

with increased plum curculio captures when compared to unbaited traps, all traps deployed in 2004 were unbaited. For both 2003 and 2004, all traps were inspected for plum curculios once, 2-3 days after the spray at petal fall of insecticide against plum curculio. Thus, results for each year show captures that occurred during a two-week period. Results show combined captures by traps in 2003 (baited + unbaited traps) and 2004.

Overwintering Study

The purpose of this study was to determine whether the presence of plum curculios in interior of blocks early in the season may be partially explained, under both sprayed and unsprayed conditions, by successful overwintering of adult plum curculios occurring within orchard blocks. I also aimed at determining, under unsprayed conditions, whether the type of weed management might be a factor influencing the extent to which adult plum curculios are able to overwinter beneath perimeter-row trees.

The study was performed from April to June in unsprayed (both in 2003 and 2004) as well as in sprayed (only in 2004) commercial orchard blocks. The study under unsprayed conditions was conducted in Cold Spring Orchard (Belchertown, Massachusetts) using two block sections (= plots), each subjected to a different type of weed management (see below). Plots were about 1 km apart. For both plots, the perimeter rows selected for this study had similar length (about 140 m) and orientation (west). For each plot there was an alleyway (about 20 m width) separating perimeter-row trees from woods (Fig. 17), which were composed primarily of deciduous trees. The grass in these

two alleyways was mowed during August/September of the preceding year. The first plot (Plot I) was subjected to weed management by means of application of herbicide and mechanical removal of weeds. Thus, at the moment of deploying emergence traps (during the early spring of 2003 and 2004), the area beneath each tree canopy was devoid of vegetation except the leaves shed by the trees in the preceding autumn. Fungicides and insecticides were also applied in Plot I, as needed, throughout 2002 and 2003. Approximately 120 rotten fruit (from the previous year) were present beneath each of the 12 perimeter-row trees (see below) in each year. The second plot (Plot II) was basically unmanaged, with no insecticide, herbicide or fungicide applied for at least six years. Thus, there was tall grass and other vegetation growing beneath tree canopies, with a small amount of rotten fruit present beneath each perimeter-row due to low fruit load.

For this study I used pyramidal emergence traps (Fig. 18) (1 x 1 m at base), made of PVC and steel screen (Pest Management Innovations, Harpers Ferry, WV). A plastic device topping each trap permitted the capture of plum curculios that, upon emergence from hibernation, walked upward on the interior surface of the trap. For both 2003 and 2004, 60 emergence traps were deployed per plot, arranged in 12 transects. Each transect consisted of five emergence traps arranged in the following manner: (a) a trap placed next to the trunk of a perimeter-row tree (denoted perimeter-row trap), (b) a trap placed in the alleyway, in close proximity to the edge of the canopy of a perimeter-row tree (denoted canopy-edge trap), (c) a trap placed in the alleyway, midway between perimeter-row trees and woods (denoted alleyway trap), (d) a trap placed at the edge of the woods (denoted woods-edge trap), and (e) a trap placed six to eight m inside the woods (denoted woods-interior trap). Traps were deployed in such a way that no plum curculios emerging within

the area covered by a trap could escape, and no plum curculios could enter a trap from the outside.

For each year, emergence traps were deployed at the silver-tip stage (on April 12 and April 14 for 2003 and 2004, respectively). Each trap was baited with one pheromone dispenser to draw plum curculios towards the capturing device. All traps were inspected for plum curculios 2-3 times per week until late June.

In 2004, I deployed 20 emergence traps in each of four commercial orchards (80 traps in all) with the aim of determining whether plum curculios may be able to overwinter inside blocks subjected to regular use of insecticide. Emergence traps were deployed in the same way that Circle traps were arranged for the orchard penetration study (i.e., four transects of five traps each).

Data Analysis

For the orchard penetration study, data were not analyzed statistically because of the several confounding variables (e.g., cultivar, tree density) that could potentially influence the outcome. For the overwintering study, I compared the number of plum curculios that overwintered beneath perimeter-row trees according to the presence vs. absence of weeds by means of a Fisher exact test.

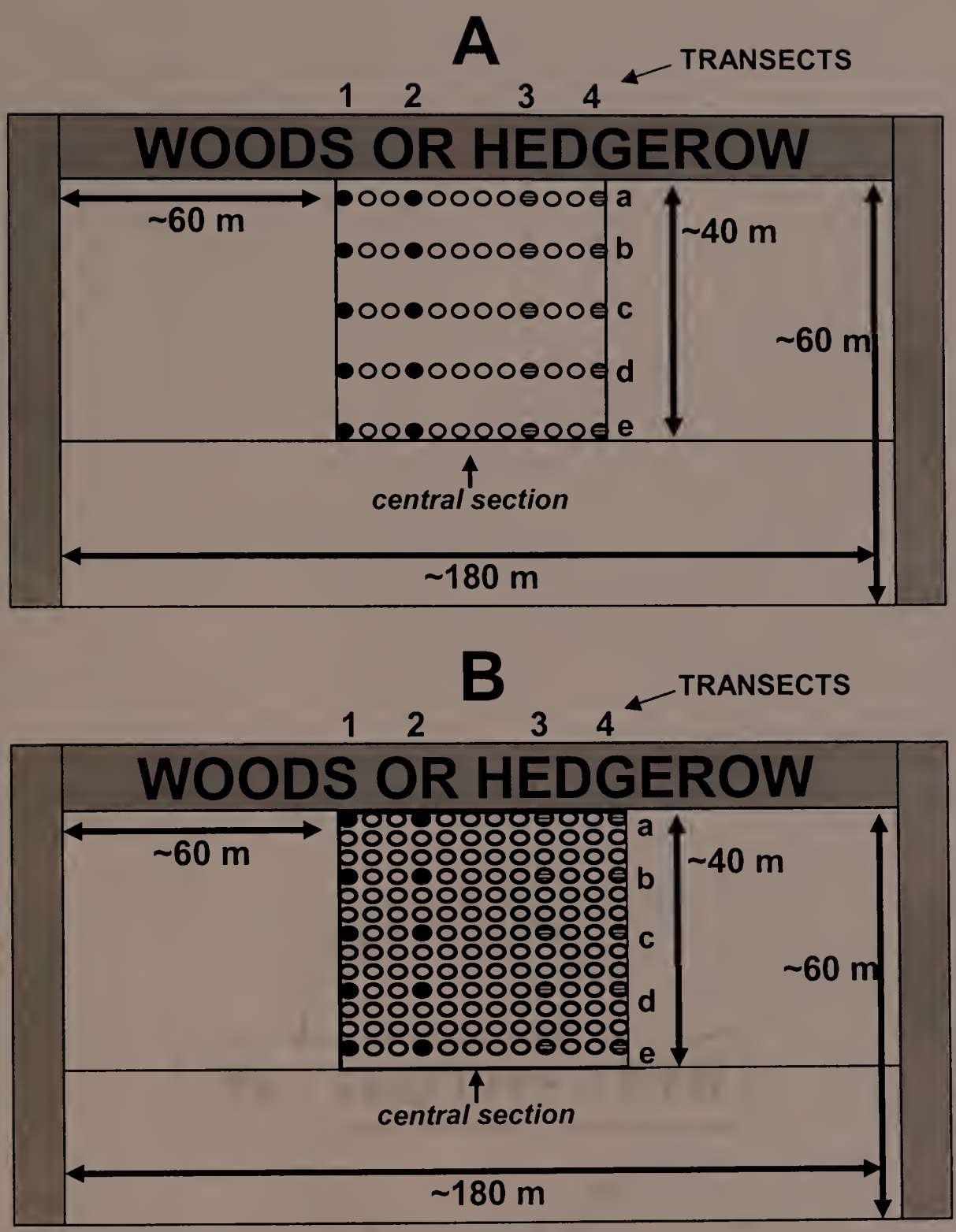

Figure 16. Layout of trap deployment in six orchard blocks having large (M.7 rootstock) trees (A), and four orchard blocks having small (M.9 rootstock) trees (B). This study was conducted in sprayed orchards in Massachusetts in 2003 and 2004. For each orchard block, 20 Circle traps were deployed, arranged in four transects each having five traps (shown as a-e). In 2003, of the 20 traps used per block, ten were baited with grandisoic acid + benzaldehyde (denoted as ●) and 10 remained unbaited (denoted as ◍). In 2004, all traps were unbaited.

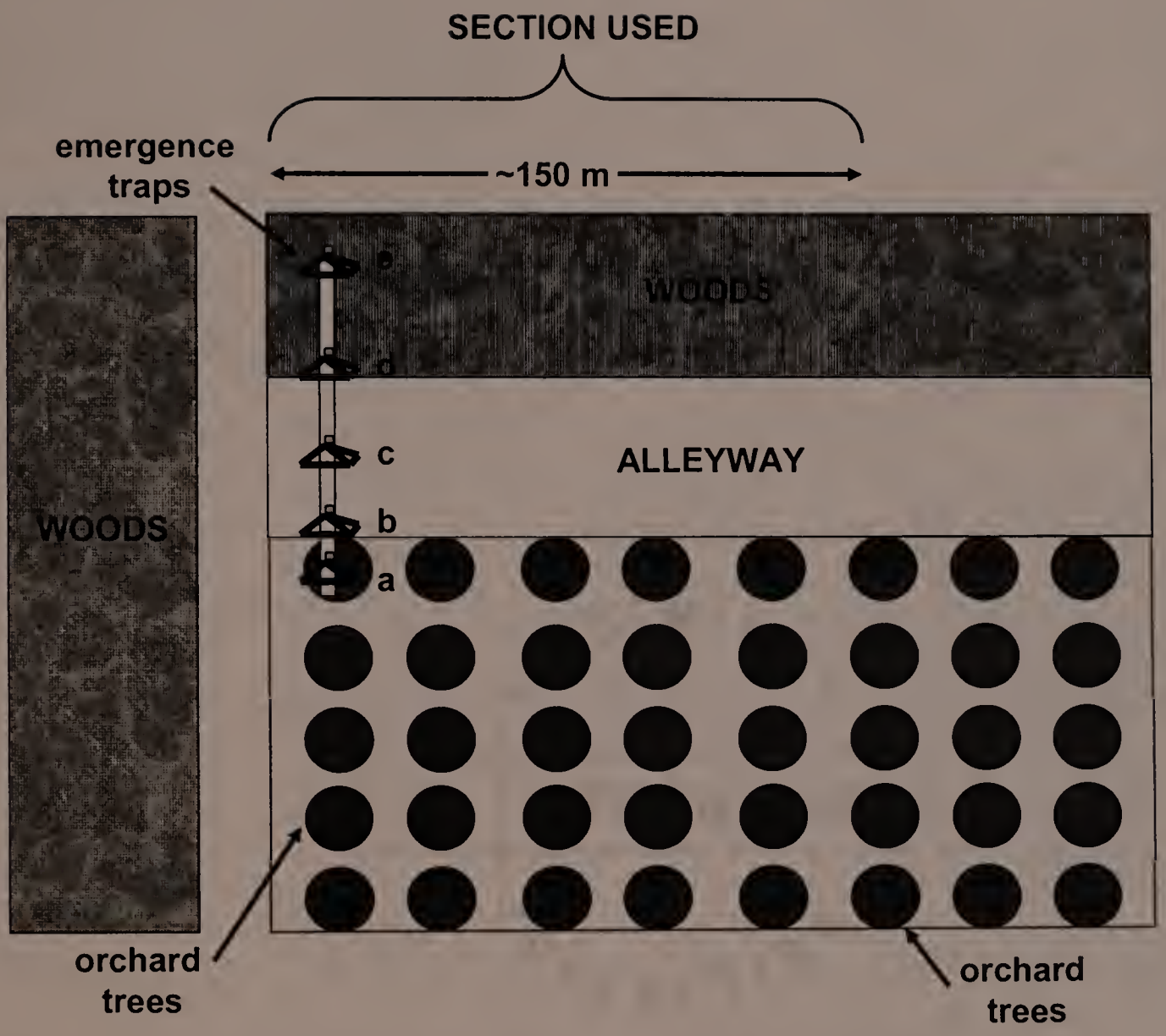

Figure 17. Trap deployment pattern used to determine extent of plum curculio overwintering in 2003 and 2004 in two unsprayed plots. Plot I was subjected to weed management. Plot II was unmanaged. On each year, 60 emergence traps were arranged in 12 transects (only one transect is shown) within each plot. Each transect consisted of five emergence traps located at different sites (denoted as a-e) along each transect.

Figure 18. Pyramidal emergence trap used for the overwintering study conducted in 2003 and 2004 in two unsprayed plots. Trap dimensions: 1 x 1 m at base.

Results

Orchard Penetration Study

Combined data from 2003 and 2004 showed that, by the time of petal fall, 61 plum curculios were captured by the 360 Circle traps deployed in the eight sprayed orchard blocks. The frequency of captures according to tree size is shown in Table 6. Plum curculios showed a propensity to be found on perimeter-row trees rather than in interior trees (63 vs. 37%) in blocks having large (M.7 rootstock) trees. In contrast, plum curculios tended to be found in interior-row trees rather than on perimeter-row trees (33 vs. 67%) in blocks having small (M.9 rootstock) trees. Regardless of tree size, a few adults were found up to 40 m inside of blocks.

Overwintering Study

Combining 2003 and 2004, 84 plum curculios were captured by the 240 emergence traps deployed in the two unsprayed plots. Data in Table 7 reveal that plum curculios were highly capable of overwintering beneath perimeter-row trees, particularly in the unmanaged plot (Plot B), although the amount of overwintering did not vary significantly between the two plots (i.e., unmanaged and managed) (Fisher exact test $P=$ 0.34). In the sprayed blocks, only 3 plum curculios were found overwintering beneath perimeter-row and second-row trees across the additional 80 traps that were deployed in the four sprayed orchard blocks used in 2004.

Discussion

I found that, by petal fall, plum curculios were more likely to be found on perimeter-row trees (as determined by captures in Circle traps) in blocks with large trees (M.7 rootstock) rather than small (M.9 rootstock) trees. I also determined that, under Massachusetts conditions, adult plum curculios were capable of overwintering inside orchard blocks, regardless of whether or not weeds were present beneath perimeter-row trees.

For the orchard-penetration study, working in sprayed blocks having relatively low plum curculio population densities and for only two weeks prevented me from obtaining large sample sizes. Yet, I determined that early-season captures of plum curculios by Circle traps in perimeter-row trees were greater in blocks having large trees than in blocks having small trees. However, there may be several factors besides tree size that may have influenced the pattern of captures. For instance, factors potentially influencing the way plum curculios perceive host quality and consequently the degree to which adults may be either attracted and arrested on an individual host plant, or induced to leave it include both the physical as well as the chemical properties of the host plant (Butkewich and Prokopy 1997, Leskey and Prokopy 2000, 2002).

Physical and chemical characteristics of host trees potentially influencing foraging behavior in the plum curculio include type of cultivar as well as vegetative and floral characteristics (Lafleur and Hill 1987). Some of these characteristics may be influenced by tree size (e.g., amounts of foliage and volatiles released, differences in phenology). Even though I was unable to randomize in this study over all possible physical and chemical factors potentially influencing my results, I believe the different

types of cues offered by trees of varying size played an important role in the outcome of the orchard-penetration study for three reasons. First, the relative influence of tree size on plum curculio foraging behavior has been addressed by Lafleur and Hill (1987), who showed in Quebec that large trees having dense foliage were able to attract higher numbers of overwintered plum curculios before fruit set than small trees, probably as a consequence of increased humidity and reduced air movement. Second, because in my study all trees within a block were of the same size coupled to the fact that trees were of the same cultivar in most blocks, I believe differences in the type and amount of volatiles emitted by trees did not differ significantly within a particular block, or at least not in a way that would determine differences in extent of penetration by plum curculios into the blocks. This study was ended by the end of the petal-fall period and thereby fruit development was incipient in all orchard blocks. Third, findings of Agnello et al. (unpublished data) show that, in Massachusetts orchards, blocks having small (M.9 rootstock) trees (primarily Gala) are more likely to present injury by plum curculio in interior trees than blocks having large (M.7 rootstock) trees (all of the cultivar MacIntosh).

That plum curculios can overwinter inside orchard blocks has been demonstrated by several researchers (Whitcomb 1929, Chapman 1938, Smith and Flessel 1968, Lafleur et al. 1987). Results found in the managed plot (Plot I) show that plum curculios are able to overwinter within orchards even in the absence of grass or accumulations of leaves, as suggested by Chapman (1938), Armstrong (1958), and Lafleur et al. (1987). The proportion of plum curculios (~54%) that were found overwintering beneath perimeter-row trees in Plot II (i.e., unmanaged) is higher than the ~20% reported by Lafleur and

Hill (1987) in Quebec, a region in which the winter months are colder than in Massachusetts. It would be important to determine whether, as we go south (e.g., Virginia, West Virginia), higher proportions of plum curculios might be able to overwinter inside orchard blocks compared to northern regions.

In Massachusetts, current approaches to controlling plum curculio involve application of a whole-orchard insecticide spray at or soon after petal fall, followed by one to three insecticide applications that are confined exclusively to peripheral-row apple trees (Prokopy et al. 2003a, Prokopy 2004). Under some circumstances, however, follow-up spraying of only peripheral rows has been shown to be insufficient for adequate orchard-wide control of plum curculio (Chouinard et al. 1992). This failure in plum curculio control could be explained in part by successful overwintering of plum curculios inside orchard blocks, in particular if there is no weed control, and by post-petal-fall penetration of plum curculios, particularly in blocks composed of small trees.

Table 6. Frequency of adult plum curculios captured by Circle traps early in the season (i.e., by the time of petal fall) according to tree size and distance from perimeter-row trees at which traps were deployed. The orchard-penetration study was conducted under sprayed (2003 and 2004) conditions in commercial apple orchards in Massachusetts.

	Small trees (M.9 rootstock)		**Large trees (M.7 rootstock)**	
Trap location	**Frequency**	**% of total**	**Frequency**	**% of total**
0 m	5	33.3	29	63.0
10-12 m	2	13.3	8	17.4
20-22 m	4	26.7	2	4.4
30-32 m	3	20.0	3	6.5
40-42 m	1	5.7	4	8.7
TOTAL	**15**	**100%**	**46**	**100%**

Table 7. Frequency of numbers of adult plum curculios that overwintered within two orchard plots in Massachusetts that differed in the type of weed management (Overwintering study). Description of trap locations is shown in Fig. 17.

	Plot A (managed)		Plot B (unmanaged)	
Trap location	**Frequency**	**% of total**	**Frequency**	**% of total**
Perimeter-row	4	30.8	38	53.5
Canopy-edge	2	15.4	5	7.0
Alleyway	0	0.0	0	0.0
Woods-edge	1	7.7	10	14.1
Woods-interior	6	46.1	18	25.4
TOTAL	**13**	**100%**	**71**	**100%**

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